

# MANUAL ON GENETIC CONSERVATION OF RICE GERM PLASM FOR EVALUATION AND UTILIZATION

Te-Tzu Chang

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#### **FOREWORD**

This manual is designed to help rice researchers to conserve, evaluate, and use existing gene-pools in the genus *Oryza*. Operations related to the scientific management of genetic resources for varietal improvement are outlined.

This manual will complement the *Manual for Field Collectors* of Rice (1972) in covering the different phases of genetic conservation.

The development of this manual was originally suggested by participants of the Symposium on Rice Breeding held at the International Rice Research Institute, September 6-10, 1971.

I wish to thanks Drs. C. Roy Adair, Peter R. Day, and S.H. Ou who reviewed the manuscript and contributed helpful suggestions and comments. Dr. Sadao Sakamoto and Miss Genoveva C. Loresto made available their experience in growing the wild taxa. Eliseo A. Bardenas, Reynaldo L. Villareal, Antonio T. Perez, and Mercedes B. Parker furnished some of the background information related to problems in registration, maintenance, and storage of genetic stocks. Bruce B. Miner edited the manuscript.

We welcome suggestions and comments that will improve the usefulness of this manual.

TE-TZU CHANG
Geneticist
and
Genetic Resources Program Leader

## Introduction

While most plant breeders recognize the importance of utilizing genetic diversity in breeding programs, much of the breeding efforts in the past has largely been based on small segments of locally adapted cultivars. Only in recent years has it become apparent that plant breeding programs with a broad genetic base could deter rapid and widespread epidemics of plant diseases and outbreaks of insect pests and thus sustain a high level of crop yield. To meet the continuously expanding needs of varietal improvement, the assemblage, evaluation, and preservation of the entire existing germ plasm are essential to more rewarding breeding efforts. This manual discusses these aspects as they relate to rice.

#### NEED FOR NEW GENES AND GENETIC DIVERSITY

Progress in plant breeding requires a continuous supply of genes or gene-complexes to meet needs that may or may not be foreseen. In rice improvement it was good luck to find a single recessive gene for semidwarfism that greatly raised the yield potential of tropical and subtropical rices. Meanwhile, with intensive cultural practices associated with progressive agriculture, the danger of serious crop losses from the ravages of diseases and insect pests becomes more imminent than before. Growing multiple crops of rice in irrigated areas greatly enhances the opportunity for a disease epidemic or a major outbreak of destructive insects.

Although rice researchers are finding and using genes or genecomplexes that will confer resistance to the major diseases and insect pests, the need for stable resistance to a shifty plant pathogen such as the blast fungus is more urgent than ever before. Similarly the need for genes that will confer tolerance to adverse environmental conditions such as water stress, cool temperatures, deep water, and problem soils, or that will enhance nutritive quality, becomes more apparent. In this respect, the researcher is often handicapped by the limited germ plasm available to him. To meet these needs, the assembling of large varietal collections, systematic screening for the desired traits, and subsequent incorporation of the genes concerned into existing cultivars is imperative. Such needs are also expected to increase in both scope and intensity when population pressure extends rice cultivation into new areas with untested soil types, different climatic patterns, new disease and insect incidences, impure irrigation water, and interactions with local flora and fauna. Some of the implications of introducing a crop into new environments are little known.

Genetic diversity in a crop species is essential to sustained levels of high productivity. Many an outstanding technological advance in crop yield or quality has stemmed from one or a few genes. Genetic uniformity within a crop is readily brought about by using the same gene or gene-complex in plant breeding and by large-scale extension of genetically related cultivars. Farmers, also demand processors. and consumers uniformity production techniques, plant height and maturity, and crop quality. As a result of narrowing genetic base of commercially important varieties, crops today are in imminent danger of serious losses from the ravages of major diseases or insects or both. When uniformity becomes the cause of genetic vulnerability, genetic diversity is the only insurance against it.

A classical example of the vulnerability of genetically uniform crop plants is the rapid spread of the Victoria blight disease on several newly released improved oat varieties in the USA during the mid-1940's when the crown rust-resistant Victoria variety was used as a common parent. It turned out that gene Pc-2 for the Victoria type of rust resistance was nearly completely linked with the gene Hv for susceptibility to the blight fungus (Helminthosporiumvictoriae). As a result, all of the Victoriaderived varieties were wiped out by a blight epidemic soon after their release.

Another recent instance of far greater magnitude in crop loss is the severe incidence of the southern leaf blight pathogen (*H. maydis*) which attacked many USA corn hybrids having the Texas male-sterile cytoplasm. The virulent T race of the fungus spread very rapidly in the Corn Belt during 1970 because the crop grown in extensive and continuous hectarage was essentially homogeneous for the susceptible T cytoplasm, though the

hybrids differed in nuclear genes. These two instances clearly show the need to reinstate genetic diversity into modern cultivars.

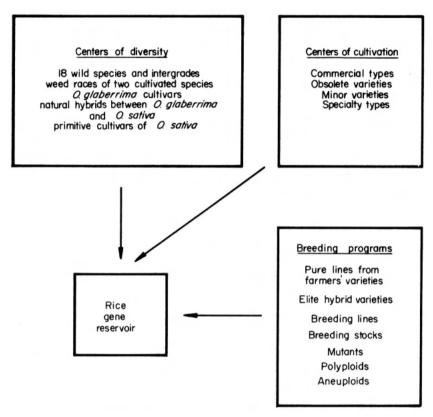
In the rice crop, the increased use of fertilizers and irrigation water favors the multiplication of disease organisms and insect pests along with the luxuriant plant growth that results from intensive cultural practices. The continuous monoculture of rice in the tropics greatly aggravates the seriousness of pest problems. Acreage planted to the semidwarfs continues to expand in irrigated areas, worldwide. All of these potential dangers call for plant breeding efforts that will continually provide a broad genetic basis in the development of new varieties as insurance against the ill effects of genetic homogeneity. Therefore, the search for useful genes should now be extended beyond a few readily accessible gene sources. Moreover, diverse gene-pools should be continually identified and utilized in breeding programs so as to provide protection against genetic changes in the prevalent disease organisms or insect pests or both.

#### SOURCES OF NEW GENE-POOLS

The entire spectrum of germ plasm existing in the rice crop is shown in Fig. 1.

New gene-pools may be indigenous or introduced from a foreign source. The following categories may characterize the various sources existing in rice.

- 1. Modern elite cultivars or high-yielding varieties. These recent products of hybridization programs have semidwarf to intermediate plant stature and high yielding ability; on the other hand, elites are often deficient in resistance to one or more major pests and in tolerance to adverse conditions.
- 2. Principal commercial varieties. Generally well adapted tothe local conditions of major production areas, these varieties have many of the desired traits, especially grain quality, for the local or export market; commercial types of recent origin are largely hybrids with characteristics intermediate between elite types and the unimproved (minor and specialty) types; those of the past have largely been selected and purified by mass or pureline selection.
- 3. *Minor varieties*. These unimproved varieties numerous within a region, probably contain many duplicates; those found in one region still contain sufficient diversity in plant and grain characteristics so that farmers continue to grow them in small



1. The full spectrum of rice germ plasm and its sources.

lots to suit different needs, e.g. maturity, cooking quality, and tolerance to adverse conditions such as deep water or drought.

- 4. Specialty types. Unusual features such as tolerance to an endemic disease, cool temperatures, or an adverse soil problem, enable these types to persist in their adapted area for a long period in spite of obvious shortcomings such as poor grain quality. Varieties reported to have special nutritive or medicinal value or those used in festive rites also belong to this group.
- 5. Obsolete types. These types were major or specialty varieties at some time in the area but their cultivation has been discontinued or is so sporadic that it is difficult to find authentic seed samples that fit the descriptions.
- 6. Breeding stocks (elite germ plasm). Breeding stocks are largely improved hybrid strains with a large number of desired

traits but fall short of being named or released because of one or more obvious shortcomings. Breeding stocks can also include  $F_1$ ,  $F_2$  hybrids, backcrosses, and multiple crosses, and their progenies.

- 7. *Mutants*. Mutants arise from mutational changes in a known genotype. Induced mutants are products of mutagenic treatments. Most of the induced mutants in rice are freakish in nature and of cursory interest. Only a few induced mutants have been found suitable for agronomic use. Occasionally a spontaneous mutant found in nature may find its way into a breeding program as parent material.
- 8. Primitive types. Cultivars that possess primitive features such as lax panicles, anthocyanin-pigmented awns, extreme shattering, strong seed dormancy, and adventitious growth of roots or tillers on the middle internodes, often carry with them other desirable characteristics such as high levels of pest resistance, tolerance to flooding, and strong competitive ability. These are generally found in less accessible areas where weed races and wild species coexist.
- 9. Weed races. These wild forms are found as companions in cultivated fields and adjacent areas where growing conditions resemble those prior to the advent of progressive agriculture. Weed races contain various intergrades between cultivated and wild forms. Farmers also harvest weed races for food; often they are grown as admixtures in a cultivated field.
- 10. Wild species. These include the 18 wild-growing taxa in the genus Oryza which can be considered as valid species (see Appendix 11). Most of the wild taxa are diploid (2n = 24) while several are tetraploid. Because of human disturbances of their adapted habitats, many wild forms have receded from cultivated sites. Some wild species are rapidly becoming extinct.

From the manner in which each of the above groups has evolved, we may postulate generally as to the genetic diversity among the members of a group from one geographic area (country or crop region), the degree of genetic homogeneity within a variety (strain) or a wild-growing population, and the agronomic value of the group in terms of productivity. From the extent of diversity within the group and of homogeneity within a population, it becomes feasible to estimate its genetic potential in breeding as a source of new traits or complexes of traits (Table 1).

Table 1. The genetic composition of different gene sources, their productivity level, and potential value in breeding.

Group	Diversity within group	Homogeneity within a strain or population	Agronomic or commercial value	Genetic potential in breeding
Modern elite cultivars	Low to moderate	Very high	Very high	Moderately high
Principal commercial types	Moderately low to moderate	Moderate to high	Moderately high	Moderate
Minor varieties	Moderately high to high	Moderately low to moderate	Moderate	Moderately high
Specialty types	Moderately low to moderate	Moderately high	Moderately low	High
Obsolete types	Moderate to high	Moderately high to high	Moderately low to moderate	Moderately low
Breeding stocks	Moderately low to moderately high	Moderate for lines; low for bulks	Most variable	Moderate to high
Mutants	Moderately low to moderate	Moderately high to high	Mostly low; few mode- rately high to high	Mostly low
Primitive types	Moderately high to high	Low to moderate	Moderately low	Low to moderately high
Weed races	Moderately high to high	Low to moderately low	Low	Low to moderately high
Wild species	Moderately low to moderate	Low to mode- rately low	Very low	Low to moderate

The foregoing generalizations also provide clues as to appropriate means of conserving the genetic variousness within a member of any group. More specific details will be discussed in the section dealing with "Maintaining the genetic composition of accessions" (p. 15).

Recent surveys on the composition of varietal collections of rice indicate that most national collections had adequate

coverage of the modern elite cultivars, principal commercial varieties, local and foreign breeding lines, and induced mutants. Nearly all national collections were deficient in minor varieties and specialty types of indigenous origin (Chang, 1972, 1975). Rather few efforts have been made to collect and conserve primitive cultivars, weed races, and wild species, and the coverage of genetic material is inadequate (Chang, 1970, 1975). While a number of countries in tropical Asia are collaborating with IRRI on the collection of minor varieties, specialty types, and primitive cultivars, it is essential that all national centers collect and preserve all segments of the indigenous rice germ plasm, much of it still existing in remote and less accessible areas.

#### GENETIC VARIOUSNESS WITHIN POPULATIONS

Samples of cultivars from farmers' fields are rarely homogenous in genetic composition. They contain varying degrees of differences in morphological or physiological aspects, some obvious to the eve while some others are not. Generally a farmer's variety can be differentiated into two or more subpopulations that are distinct in one or more recognizable traits. In addition, a small number of obvious off-types are often found in the seed sample. The extent to which an accession or sample can be separated into sub-populations depends on the existing degree of recognizable grades in the trait or on the refinement of a technique by which the trait can be further resolved into component traits. For instance, an accession is initially differentiated into two sub-populations by the presence or absence of purple pigments in the basal leafsheaths of the young plants. By inoculating the plants in each group with a known strain of the bacterial leaf blight pathogen, each sub-population may be found to contain many susceptible plants as well as a few that are moderately resistant. We now have four groups. When several bacterial strains are used to inoculate progenies of the four groups, a further division into many lines based on reaction types is often feasible. If such lines are further tested for gelatinization temperature of the grain, a resistant line may be found to contain both "low" and "intermediate" individuals. Therefore, the differentiation of an accession into many subpopulations is a dynamic process, depending on the number of traits being tested and the degree of resolution of each trait. If we have 10 tests available for such a purpose, it would not be

difficult to resolve one farmer's variety into hundreds of strains or biotypes.

Likewise, modern varieties of hybrid origin are not necessarily pure lines. While the degree of genetic purity is related to the amount of pedigreed selection and seed purification applied to the variety, even highly purified varieties will show signs of segregation when grown at a new location or in a different crop season. Much of the segregation will show up as variability in the date of heading, adult plant height, panicle exsertion, and panicle number, all of which result from an interaction between genotypic variants in the variety and a specific environmental condition (daylength, air or water temperature, or water supply). An example of "environmental segregation" (Bennett, 1970) found under different photoperiods in the F<sub>8</sub> population of IR20 is given in the IRRI annual report for 1970 (IRRI, 1971, p. 218).

Inherent variousness within an accession should be preserved and handled by the genetic stock officer in the interest of genetic conservation in a manner different from that used in conventional plant breeding. Since it is impossible to maintain the tens or hundreds of strains separable from each accession in a highly purified state, no effort should be made to select for certain types at the risk of losing others. Therefore, the accession should be maintained as a bulk much the same as collected from the field, so that its genetic composition will not be markedly altered. Obvious off-types, however, should be removed whenever feasible.

On the other hand, elaborate research in genetics, pathology, entomology, physiology, or biochemistry requires the use of genetically pure strains so that experimental findings can be repeated and readily verified. Such studies demand the maintenance of pure lines and periodic checks on their genetic purity.

When pure strains of special merit are identified from an accession by the pathologist or the entomologist, they should also be given to the genetic stock officer for preservation. An additional number or code such as -1 or -A to the original accession number should be given to distinguish it from the original bulk sample. Additional numbers or codes may be added if further division is warranted by other tests. Such designation is similar to the pedigree system in plant breeding.

While the isolation of pure strains and the maintenance of the

bulk population may appear to be efforts in different directions, they are actually complementary facets of conservation and utilization programs.

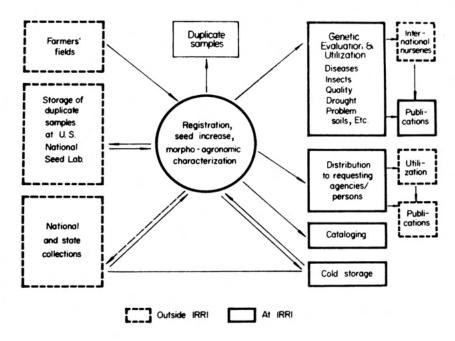
#### GENETIC CONSERVATION VERSUS EVALUATION AND UTILIZATION

Genetic conservation aims to preserve all of the genetic variousness that is available in a population. The term implies a minimum amount of selection or removal, except for obviously duplicate accessions and apparent off-types. Conservation is best insured by a minimal number of cycles of seed rejuvenation under an environment as similar as possible to the native habitat of the population. On the other hand, evaluation and utilization necessitate the identification and isolation of obviously useful genotypes, involving selection and purification in the process. Splitting of original stocks into pure lines, and repeated seed-increase cycles, are adjuncts of evaluation and utilization. However, the two processes are not mutually exclusive and can be operated in a complementary manner. Conservation provides the inputs while evaluation and utilization make conservation efforts worthwhile.

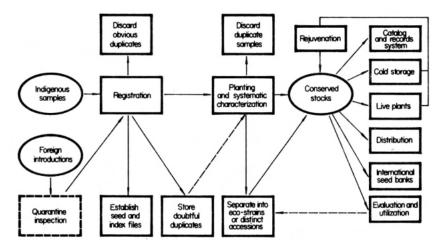
In operational terms it is desirable to plan for conservation at the initial stage of seed increase. During preliminary evaluation a judicious roguing of obvious off-types in the original population is about all that is needed, so that the genetic composition of the sample remains unaltered. When a larger seed stock is obtained from the initial seed increase, a representative sample should be sent to a seed bank for long-term storage. The national center will then initiate efforts in evaluation and utilization that will involve selection and purification. Because of the unknown genetic potential in the not-so-pure original sample, it is necessary for the national center to store the remnant seeds while the evaluation process is being carried out, so there will be an opportunity to retrieve the desired segment of genetic variousness in the original sample when needed. The sample stored at the seed bank further insures the availability of the original stock.

#### PROCEDURE IN GENETIC CONSERVATION AND UTILIZATION

The steps adopted at IRRI to assemble, characterize, evaluate, utilize, and preserve 35,000 genetic accessions of rice are illustrated in Fig. 2. The procedure for collecting rice samples



2. Flow chart showing the different operations at IRRI in acquiring, increasing seed, cataloging, preserving, evaluating, and utilizing rice germ plasm with the collaboration of other agricultural research centers.



**3.** Flow chart outlining the necessary steps in the processing of rice seed samples for registration, seed increase, characterization, evaluation, utilization, and preservation at a national rice research center.

has been discussed in the Manual for Field Collectors Of Rice (Chang *et al.*, 1972). Details on characterization, evaluation, and conservation will be discussed in the following sections.

While various national rice research centers are expected to organize characterization, evaluation, and maintenance programs to suit different national needs, the sequence in the handling of genetic stocks will primarily follow that used at IRRI. The procedure illustrated in Fig. 3 may be useful as a reference for national centers.

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# **Characterization and Maintenance of Genetic Stocks**

The initial planting of collected samples can fulfill the following functions: increasing seed stocks, morpho-agronomic description by systematic recording, elimination of obviously duplicate samples by comparing morpho-agronomic traits, and preliminary evaluation and selection of suitable accessions for more intensive evaluation and for preservation. Efforts in characterization should eventually lead to a system of recording and storing useful data that can be readily retrieved and made available to other workers.

#### OBJECTIVES AND REQUIREMENTS OF SYSTEMATIC DESCRIPTION

Systematic description provides the basis for 1) characterizing cultivars or breeding lines in the national or regional interest, 2) differentiating between accessions with identical or similar identifying accessions having characteristics, 4) classifying commercial varieties on sound criteria, 5) developing interrelationships between or among traits and between geographic groups of cultivars, and 6) estimating the extent of variousness within the varietal collection. Most of the above objectives are closely related to researches in genetics, breeding, agronomy, entomology, plant pathology, physiology, cereal chemistry, and plant biochemistry. Systematic description often leads to a more efficient use of germ plasm in the collection. Data recorded from the accessions of a national collection generally lead to the publication of a varietal catalog or bulletin.

Systematic description requires the planting, cultural management, and recording of a large varietal collection under uniform conditions so that differences in recorded traits represent true varietal characteristics expressed under such environmental

conditions. The traits to be recorded should have value in taxonomy, agronomic utilization or, in some instances, academic investigations. The traits should have reasonably high heritability so that the findings can be verified. In reporting the recorded information, it is essential to include a pertinent description of the climatic conditions, soil type and chemical properties, cultural practices, and planting dates at the station site so that readers may make meaningful comparisons.

#### PLANTING AND MULTIPLICATION OF SEED STOCKS

Seed increase is a necessary evil following collection or introduction. Not only is it costly and time consuming, it also involves the risk of losing accessions through poor adaptation or disease and pest damage, introducing admixtures through contamination or error, and altering the genetic composition of the original population through conscious (human) or unconscious (natural) selection. Therefore, it is essential to increase seed stocks sufficiently in one cycle so that the harvest will serve different uses in evaluation, distribution, and storage. Repeated seed increases in small plantings should be avoided.

For seed multiplication and for preliminary evaluation, a favorable environment similar to the native habitat of the cultivars or wild taxa should be provided to the extent possible so as to minimize natural selection due to unfavorable environmental factors. Similar consideration should govern the choice of cultural practices and water management.

The number of plants of each accession to be grown depends on the need for increased seed and the experimental facilities available. Since repeated cycles of seed increase are not advisable, it is best to plant enough seeds for one efficient cycle of seed increase. Because of wide spacing and because some of the collected samples may not be well adapted to the experiment station environment, a yield no greater than 3,000 kg/ha should be anticipated.

Forty single-plant hills in a transplanted culture or two 5-meter drilled rows is the minimum for each accession; 60 to 100 plants is about optimum.

The researcher should use one seedling per hill in a transplanted culture, or more widely spaced drill-planting in a directseeded culture, in order to facilitate roguing and sorting. Complete fertilizers should be applied to insure adequate plant growth, but at a rate that will not induce serious lodging. A relatively wide spacing between rows will be helpful in separating lodged plants from one another. Other cultural and plant protection practices should be designed so that sufficient healthy seeds can be obtained from the planting.

An experiment plan and copies of field notebooks should be prepared prior to seeding. Details on this operation are discussed in the section dealing with "Method of recording plant characters and growth stages" (p. 18).

When seeds come from a foreign source, treat the seeds according to the prescribed plant quarantine measures. During the crop season, watch out for signs of seed-borne diseases or nematodes. Complete plant protection measures are generally necessary for strains of unknown genetic potential. When disease infection or insect infestation appears on an accession, the worker can be reasonably certain as to the susceptibility of the variety. But inference about varietal resistance because of a disease - or insect-free condition is unreliable, since escape from infection or infestation by chance is common in field plantings.

Before seeding, it may be useful to test the germination of accessions that were collected when premature or overripe, or of those coming from storage bins or market places. When only a few viable grains of an accession are available, grow them in pots or special plots so that the preliminary seed increase and evaluation can be carried out later on an adequate scale.

When a seed sample came from an outside source, be sure to divide it and save the second portion. The remnant seeds are often needed for another planting when the first effort fails, or for future use.

Rows or plots of one or two standard varieties should be planted at regular intervals to serve as controls. Separate adjacent plots with a blank row. This will help in sorting out lodged plants belonging to different plots.

Accessions belonging to the same crop season or maturity group should be planted together at one date of seeding. Accessions that are suspected to be duplicates should be grown side by side to facilitate comparison.

For accessions that fail to germinate in the seedbed, treat the remnant seed with an appropriate fungicide and replant as soon as possible; and if that fails, try to obtain another seed sample from the same source.

During the process of growing plants, attention should be given to minimize natural cross-pollination, contamination, and erroneous labelling. Similarly, contamination at harvest time, erroneous recording, and subjective selection within a naturally heterogeneous population should be minimized. Practices and measures to attain such objectives are described in the section dealing with "Maintaining the genetic composition of accessions" (p. 15).

The weight of grains harvested from each plot should be recorded as it will give some indication of yielding ability.

The facilities needed for the above operations are common to those of an ordinary experiment station engaged in rice breeding and agronomic trials. Some of the basic requirements in experimental facilities are fields of regular shape, adequate size, and uniform fertility; dependable water supply; plots provided with independent irrigation and drainage facilities; equipment for seed processing and storage; machinery for field operations; and a supply of agricultural chemicals and other experimental materials (stakes, bags, labels, stationery, etc.). To facilitate year-round planting of rice, a series of irrigated nursery beds protected by wire nets is most useful in minimizing rat and bird damage. For stations with cooler climates, glasshouses are needed. Dark rooms will be useful in inducing photoperiod-sensitive cultivars to flower within a reasonable period of time.

#### MAINTAINING THE GENETIC COMPOSITION OF ACCESSIONS

When the original sample contains detectable variousness or separable sub-strains, it is important to first distinguish between obvious contaminants (off-types) and inherent variants within a population. Obvious off-types or contaminants, such as awned and bold-grained plants in an awnless and slender-grained population, should of course be removed. In an inherently heterogeneous population, the variants will appear when the different but related genotypes interact with the environmental conditions prevailing at the planting site. The worker should maintain the variants either in bulk or as separate sub-populations until he finds a valid basis to split the population into strains or to combine a strain with another related accession. Either splitting or pooling can be more effectively carried out when characterization and evaluation are largely completed.

The following operations will be helpful in maintaining the original genetic composition of rice accessions:

- 1. Preserve several grains of the original seed sample in a seed file and arrange the seed sample as well as the field collection records according to the accession number so that when any doubt should arise about the accession concerned, the planted material may be readily checked against the original seed sample and field collection record.
- 2. Keep the seedbed and the field free from volunteer plants (which generally arise from dropped seeds in the previous planting) by alternate drying and wetting of soil, thorough soil preparation, frequent roguing, and allowing a sufficient period of dry fallow between crops. In temperate regions, a rotation system involving other crops may be used.
- 3. Insert a standard variety at regular intervals, such as every 20th plot, to serve as a check.
- 4. Use the same plot number and label in the seeding, transplanting, and harvesting processes to minimize errors; keep a field map of the plot layout in the field.
- 5. Use consecutive numbers for plots grown on the station within the same crop season; never use the same plot number twice in one season.
  - 6. Rogue off-types several times during the growing season.
- 7. Use tags or other labelling devices to identify plants belonging to two or more sub-populations of the accession.
- 8. Use stakes and ropes to separate a lodged plot from its adjacent plots.
- 9. Harvest the sub-populations separately and give the appropriate identification numbers.
- 10. Clean the thresher or winnower after processing each accession or plot.
- 11. Use new or carefully cleaned containers (cloth sacks, paper bags, metal cans) in harvesting, drying, and storing seeds.
- 12. Check every labelling or recording operation for possible errors.
- 13. Prepare all records in duplicate; have a second person verify labels and records; file duplicate copies of record books in rat- and fire-proof cabinets.

When an accession has been subject to selection, splitting, or pooling, it needs to be re-identified by its seed source, plot number, and a notation on the process (by a standardized suffix

to the accession number). Example: IRRI Acc. 1107-PP denotes a blast resistant strain of Acc. 1107 selected and purified by the Plant Pathology Department. Similarly, variants of a desirable type should also be saved and given an appropriate designation.

#### PLANT AND GRAIN CHARACTERISTICS FOR DESCRIPTION

The principal morpho-agronomic characters to be systematically recorded at different growth stages are as follows. For genetic stock officers who have limited help and time, the most essential traits are shown in italics:

Young seedlings — rate of seedling emergence, *leaf color*, *seedling height* (at 4-leaf stage, shortly before transplanting).

Juvenile plants — rate of tillering, leaf length, leaf angle, leaf color (intensity of green as well as anthocyanin pigmentation), color of basal leafsheaths.

Plants around maximum tillering — tiller number; leaf length, width, angle, hairiness, and *color*; color and length of ligule; color of collar; color of auricles; color of leafsheath.

Plants at and shortly after heading — panicle number; angle of inclination of tillers; leaf angle of the uppermost blade; date of full heading, and uniformity of heading among plants in a plot; internode color; sheath pulvinus color; culm length or plant height; flagleaf angle and length; stigma color.

Plants during the grain-ripening stage — panicle type; panicle exsertion; panicle length; culm diameter and strength; awn presence and color; color of apiculus, lemma and palea, and sterile lemmas; panicle fertility; panicle threshability; leaf senescence; date of maturity.

Grain features after harvest — *length, width,* and L/W ratio; color of apiculus, lemma, and palea; hairiness of hull; *color of seedcoats; endosperm type* (waxy or common); embryo size; *100-grain weight, chalkiness of endosperm* (white belly, white center, and white back); amylose content; gelatinization temperature; protein and lysine contents.

Natural incidence of diseases and insects at all stages — kind, severity, stage of growth.

Tolerance to adverse conditions at all stages — flooding, wind damage, drought, cool temperatures, nutritional disorders, or adverse soil factors.

Consult IRRI Technical Bulletin 4, "The morphology and varietal characteristics of the rice plant," (Chang and Bardenas,

1965, p. 17-25) and the FAO Handbook of Plant Introduction (Chang, 1974) for standard definition of plant and grain characters; and the IRRI variety catalog (IRRI, 1970) for the appropriate time to make measurements, counts, or observations. Suggested sample sizes for different traits are given in the catalog. A sample sheet from the catalog is given as Appendix II.

Information on systematic description being carried out in Taiwan, Japan, and India is given by Taiwan Agricultural Research Institute (1964), Ito (1965), and Indian Council of Agricultural Research (1971), respectively.

#### METHOD OF RECORDING PLANT CHARACTERS AND GROWTH STAGES

For every planting or experiment, the researcher should prepare an experiment plan and data records. The experiment plan (planting plan) should identify the title of the experiment, the year or season or both, station and location, the accessions to be grown and tested, and information on experimental design and cultural practices. Field data should be entered in field books prepared in duplicate or triplicate and identified by year, season, experiment number and title, and researcher-in-charge. Columns in the book should include plot number, accession number and name, seed source, origin, and the items of the data to be recorded. Enter the information in waterproof ink. Pages in the book should be numbered. After field planting is completed, record the dates of seeding and of transplanting, site (field number), and a map of the experimental plots.

To facilitate the recording of dates, attach a list to the book that gives the number of days from seeding (DAS) or from transplanting (DAT) corresponding to the date given by month and day. For instance, when seeding is made on June 1, the following list will be useful.

Month and day	DAS
July 1	31
August 1	62
September 1	93
October 1	123
November 1	154

Since a large number of accessions is generally involved, the researcher will find it efficient to record certain traits in coded

numbers or letters. An increasing scale of numbers (from 0 to 9) is generally used to indicate the relative ranking of a trait from little to much, or from desirable to undesirable. The decimal coding system should be well designed and continually used from year to year.

A typical coding scale follow	A	typical	coding	scale	follows
-------------------------------	---	---------	--------	-------	---------

Leaf blade color		Disease reaction
Blank or dash ( — )		Blank or dash ( — )
None		Absent
Pale green	•	Very resistant
Chlorotic stripes R	: ₹	Resistant
Yellowish green	ſ	Moderately resistant
Green	ſ	Low intermediate
Dark green N	ИΚ	Intermediate
Purple margins	•	High intermediate
Purple trace	•	Moderately susceptible
Purple S	: {	
Dark purple	ι	Susceptible Very susceptible
Present		
Mixed		Mixed
	Blank or dash ( — )  None Pale green Chlorotic stripes Yellowish green Green Dark green Purple margins Purple trace Purple Dark purple Present	Blank or dash ( — )  None Pale green Chlorotic stripes Yellowish green Green Dark green Purple margins Purple trace Purple Dark purple Present

<sup>\*</sup> Or missing plot.

Examples of coding plant characters are described in the IRRI variety catalog (IRRI, 1970) and "Standard system for scoring rice experiments" (IRRI, 1975). Additional details are given in the section pertaining to "Data recording, processing, and retrieval" (p. 43).

Data for accessions in one experiment or a series of experiments should be taken by the same team of workers so that uniformity in observation and rating is assured. Complete the recording of a trait or traits in one experiment within the same day or the shortest period possible. For quantitative traits, use the appropriate measuring devices.

Because rice cultivars vary appreciably in their vegetative growth duration and in growth behavior, the recording of plant characteristics, responses to environmental conditions or controlled treatments, and reactions to pathogens or insects should be made both on a chronological scale (as the number of days from seeding or transplanting) and on a physiological scale (the appropriate growth stage). The chronological records are important to the local workers when seeding is made each year

(or season) on a fixed date or in a certain period. The growth stage on the physiological scale becomes more critical when a researcher wishes to compare varieties that differ greatly in their growth duration or growth pattern. For instance, an earlier maturing variety would show bacterial leaf blight infection on its flag leaves at an earlier date while a later maturing variety would appear disease free on the same date. In another respect, when early maturing varieties do not have a lag vegetative phase, the chronological date after maximum tillering is not comparable to that of the late-maturing or photoperiod-sensitive types which have a pronounced lag phase.

When time and manpower permit, the growth stages can be recorded on a decimal scale which is further divided into the principal (1-digit) and the secondary (2-digit) stages (Zadoks, Chang, and Konzak, 1974). The stages and descriptive details are in Table 2.

#### Elimination of duplicate accessions

Duplicate accessions are extra samples of the same variety (or strain) or nearly identical variants of a variety. Duplicate accessions arise when one variety has spread to several locations, resulting in 1) morphologically identical varieties with slightly different names coming from adjoining areas or countries, 2) varieties with quite different names but essentially identical morpho-agronomic features, and 3) eco-strains having the same name but which can be differentiated by their physiological responses to the prevailing physical or biological environment. On the other hand, in many instances accessions with the same name are entirely different from the original variety after seeds have changed hands and repeatedly multiplied, resulting in changes in name or genetic identity. For these reasons the need to keep a seed file of all original seed samples received is clear.

For countries having different dialects, a list of synonyms should be compiled to facilitate the identification of seed samples at national and international centers. At an international center the genetic officer should have a working knowledge of different linguistic designations such as early, late, glutinous, lowland, upland, red, white, and scented.

One interesting example is a Taiwan indica variety, Bir-me-fen (meaning White Rice Powder), which originally came from the

Table 2. A decimal code for the principal and secondary growth stages of rice.

1-digit code	Principal growth stage	2-digit code	Secondary growth stage
0	Germination	00	Dry seed
		01	Start of water imbibition
		03	Seed fully imbibed
		05	Radicle breaks
		07	Coleoptile appears
		09	Primary leaf appears at
			coleoptile tip
1	Seedling	10	Primary leaf half-way
			through coleoptile
		11	Primary leaf unfolded
		12	Two leaves unfolded
		13	Three leaves unfolded
		14	Four leaves unfolded
		15	Five leaves unfolded
		16	Six leaves unfolded
		17	Seven leaves unfolded
		18	Eight leaves unfolded Nine leaves unfolded
0	Tillering	19 20	Main shoot only
2	Tillering	20 21	First primary tiller appears
		23	Primary tillers develop
		25 25	Secondary tillers develop
		27	Tertiary tillers develop
		29	Maximum tillering stage
3	Stem elongation	30	Leafsheath clongation (during
· ·	Green elenganen	00	vegetative — lag phase)
		31	One node detectable — panicle
			primordium initiated '
		33	Two nodes detectable — panicle
			primordium visible
		35	Three nodes visible — panicle branches visible
		37	Four nodes visible — spikelets visible
		39	Flag leaf ligule/collar just visible — meiosis
4	Booting	41	Flag leaf sheath extending — early boot stage
		43	Boots just visible swollen —
		45	mid-boot stage Boots swollen — late-boot stage
		47	Flag leaf sheath opening
		49	First awn just visible
5	Heading	50 } 51 }	First spikelet of panicle just visible {
		<sup>52</sup> <sub>53</sub> }	1/4 of panicles headed {N
		54} 55	$\frac{1}{2}$ of panicles headed $\frac{N}{S}$
		56 57 <b>}</b>	¾ of panicles headed {N

#### table 2 continued

				_
		58 59}	Heading completed	{ N S
6	Flowering	<sup>60</sup> <sub>61</sub> }	Beginning of flowering	{
		62 63}	¼ of spikelets flowered	{
		<sup>64</sup> <sub>65</sub> }	½ of spikelets flowered	{\s^N}
		66 67 }	% of spikelets flowered	{ N
		68 69	Flowering completed	{ <b>^</b> N
7	Milk stage	71	Caryopsis watery - birefrigence of starch granules begins	,
		73 75 77 79	Early milk stage Mid-milk stage Late-milk stage Endosperm pasty white	
8	Dough stage	81 85 89	Early dough stage Soft dough stage Hard dough stage	
9	Ripening stage	90 91	Caryopsis hard	{ !
		92 93}	Caryopsis fully ripe	{!
		94	Overripe, straw dead	
		95	Seed dormant	
		96	50% seeds dormant	
		97	Seed no longer dormant	

Supplementary code for transplanted rice only:

T - Transplanting and recovery

T1 - Uprooting T3 - Rooting

T5 - Intermediate note T7 - Recovery of shoot

T9 - Resumption of leaf growth

N - Non -synchronous crop

S - Synchronous crop

China mainland. During World War II, Japanese soldiers took Bir-me-fen to Malaya where it became a major variety from 1945 to 1964 and was known as Pebifun. Seeds of Pebifun further spread to neighboring countries in Asia and later to nations in originating Africa. Four accessions from Bir-me-fen entered the IRRI collection: Bir-me-fen (from Taiwan), Pe-befun (from Malaya via the Philippines), Pebifun (from Malaya via the USA) and Pe-bi-hun (from Malaya via India). Through repeated cycles of growing and purification at different sites, the four accessions now differ appreciably in culm length (from 104 to 126 cm) and in maturity (from 105 to 137 days), so as to qualify as four eco-strains of one cultivar. Other plant and grain characteristics are identical, however.

Although the elimination of obvious duplicate accessions is a tedious process, it is necessary so that maintenance work can be efficiently handled without the burden of carrying the apparently duplicate entries. The process should begin at the time of registering a collected sample or an introduction. Check the variety name and seed source against the record of the registered accessions. Look for similar names or seed source or both in the accession cards or books. Then compare those features given in the field collection sheet or other records against those of the registered accessions. Grain features of the newly acquired entry or entries can be readily compared with those of the old accession in the seed file. Grain length, width, and shape; hairiness and color of glumes; and chalkiness of endosperm are useful though not always conclusive criteria in the initial process of identification.

Discard the obviously duplicate sample or samples when the variety name and origin are the same. If in doubt, place the sample in question in storage until a time when other means of comparison are available. If one has difficulty in making a decision, grow the new accession along with the registered one in adjacent plots for detailed comparison.

When two probably duplicate samples are grown in the field, compare plant height, pigmentation of plant parts, heading date, panicle number and characteristics, and several morphological features of the plant to determine if differences exist. Other features useful in comparison are disease reactions, amylose content and gelatinization temperature of the endosperm, and the number of days from first heading to full heading. Apply any other biochemical tests whenever feasible. Discard the obvious duplicate or duplicates, when identified in such a manner. When two samples are distinctly different, both will be accepted and maintained as separate accessions under two accession numbers though they may share the same name. In such cases, the different seed sources or origins indicate the reason for keeping two or more accessions under one name.

Eco-strains pose a different situation since they show subtle

but distinct differences in physiological characteristics such as flowering time and adult height, and disease or insect reactions. At the national center, new ecotypes should be preserved or discarded on the basis of their individual merit, that is, keep only those of obvious or potential usefulness to the national need. However, the national center may find it advantageous to send the distinct eco-strains to the international seed bank where they will be preserved along with the original variety.

A hypothetical example of separating five samples having similar or related names into duplicate accessions, distinct accessions, and eco-strains is given in Table 3. By comparing the 14 traits recorded, the researcher may conclude that sample 3605 and sample 5739 are duplicates of sample 407 (which was maintained by the national center). Sample 4703 is morphologically and physiologically distinct from sample 407

Table 3. A comparison of five samples having similar or related names.

Sample No.	407	3605	4702	5739	6558
Name <sup>a</sup>	Gendjah Banten	Gendjah Banten 11	Gendjah Banten	Gendjah Banten	Gendjah Banten
Seed source <sup>b</sup>	3-1	3-4	3-5	3-6	4-1
Origin <sup>c</sup>	3	3	3	3	3
Basal leafsheath	green	green	purple	green	green
Blade length (cm)	78	77	82	76	74
Blade width (cm)	1.6	1.5	1.9	1.5	1.3
Culm length (cm)	138	137	143	139	128
Culm number	26	24	22	25	20
Flagleaf angle	horizontal	horizontal	drooping	horizontal	horizontal
Panicle length (cm)	26	27	30	28	25
Panicle type	compact	compact	open	compact	compact
Grain length (mm)	8.3	8.4	9.4	8.2	8.1
Grain width (mm)	2.9	3.0	3.1	3.0	2.8
Lemma color	straw	straw	brown furrows	straw	straw
Maturity (days)	179	182	190	180	149
Blast reaction	MR	MR	S	MR	R
Bacterial leaf blight reaction	MS	MS	R	MS	MR

<sup>&</sup>lt;sup>a</sup>Fictitious names. <sup>b</sup>Code for country and station (or district). <sup>c</sup>Code for country

though the two samples have nearly identical names. Sample 6558 appears to be an eco-strain of sample 407 after having been grown and selected for adaptation in a different country.

In actual operations, however, the genetic stock officer would often find it more difficult to determine whether the newly received samples are duplicates or eco-strains or distinct accessions. Inherent genetic variousness within a sample and overlapping variation of a trait between samples will complicate the process of differentiating among samples. This is one area where additional biochemical tests such as the comparison of isozymes are sorely needed.

CULTURE AND IDENTIFICATION OF GENETIC TESTERS AND WILD SPECIES

#### Genetic testers

Genetic testers or gene-markers are generally weak in growth and poor in yield. Most testers are highly susceptible to diseases and insects. Special care in culturing the testers, beginning from seed treatment prior to seeding, followed by complete plant protection measures, and ending with the bagging of panicles at flowering, should be practiced.

During the growing season plant characteristics of gene markers should be checked frequently to insure true identity. Descriptions of the common testers are given by Chang and Bardenas (1965). The person using the genetic testers should communicate with the worker who first identified and supplied the gene markers for the characteristic features of each marker concerned.

Some of the testers such as chlorophyll-less mutants and aneuploids need to be maintained in the heterozygous condition by frequent backcrossing to a parent. Partially sterile testers need to be bagged to prevent cross-pollination. Pedigree records should be kept on each tester.

#### Wild species

The culture of wild species presents more problems than that of the genetic testers. The seeds generally have very strong and long dormancy. The seedlings are slow growing and therefore vulnerable to careless handling. The wild taxa are generally susceptible to the bacterial and virus diseases and to the leaf-hoppers and planthoppers. The seed samples are generally heterogeneous in nature, requiring the growing of sufficient

plants to represent the sample and the maintenance of the population in bulk form. Other difficulties encountered in the culture and maintenance of the wild taxa are low adaptiveness to different environments, strong photoperiod sensitivity, high frequency of out-crossing, low spikelet fertility, extreme shattering, and low rate of seed increase per generation.

To facilitate the culture of wild forms, we suggest the following measures.

- 1. Break dormancy of freshly harvested seeds by heat treatment (50°C-54°C for 5 days) and by dehulling and scratching the pericarp near the embryo.
- 2. Treat dehulled seed with organo-mercurial solution, wash; germinate in petri dishes.
- 3. Plant germinated seed in a flat or tray containing moist, fine soil; grow young seedlings under partial shade.
- 4. Transplant at 3-leaf stage (about 30 days from seeding); water plants carefully (plants are slow growing); apply fertilizer in small amounts.
- 5. Many strains are strongly photoperiod sensitive and may require short-day (10-hour) treatment to induce panicle initiation.
  - 6. Protect plants against leafhoppers and virus diseases.
- 7. Bag newly emerged panicles to collect easily shattered seeds; use ventilated bags to facilitate anther dehiscence.
- 8. Cut back the plants at about the midpoint of their culms after the first harvest to induce a ration crop, since one planting usually does not provide large quantities of seeds.
- 9. Keep seeds coming from different plants in separate lots, since wild taxa are heterogeneous and heterozygous.

The classification and nomenclature of the various wild taxa of the genus *Oryza* are still in a state of flux. In recent years several taxa have been removed by agrostologists from the genus (Chang, 1964, 1970) while a few others have been renamed (Clayton, 1968). We found that many of the wild taxa received at IRRI were mislabelled and that they require re-identification and proper designation.

To facilitate identification, we have devised a key for classifying and naming the valid taxa in the genus (Appendix I).

Intergrades of hybrids between two species pose problems for identification and nomenclature. In tropical Asia, natural hybrids between *O. sativa* and either one of its wild relatives,

O. nivara and O. rufipogon, are more commonly found in nature than are typical samples of either wild taxon. Some of the hybrids showed introgressive hybridization with either one of the parents. We tentatively call these samples the "spontanea" forms of O. sativa (Chang, 1975). Hybrids between O. sativa and O. glaberrima have appeared in samples received from West Africa.

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### **Evaluation and Utilization**

Newly collected seed samples, either domestic or foreign, warrant evaluation by competent workers before being used in a breeding program. Such new genetic material generally offers potential value because of one or more characters rather than for direct use as a commercial variety.

The incorporation of the semidwarf plant type into tropical rices has markedly raised the yield potential. Similarly, the range of geographic adaptation has been greatly extended by the use of photoperiod-insensitive and early maturing genotypes. Further progress in varietal improvement will be largely aimed toward increased and stabilized productivity through varietal resistance to physical or biotic components of environmental stress and toward upgrading the physiological production potential of the crop.

Promising sources of genes controlling resistance to pests or tolerance to adverse physical environments are more likely to be found in minor varieties, specialty types, or primitive cultivars. Fortunately, the spread of high-yielding varieties has been slow in areas where such types are most likely to be found. Therefore, these potentially valuable gene pools are still available for use by riceresearchers.

Systematic evaluation for any trait is a costly and timeconsuming process. Therefore, the researcher should have welldefined goals and procedures before he begins the tests.

#### OBJECTIVES AND TYPES OF SYSTEMATIC EVALUATION

Evaluation of collected varieties or samples can be made in different ways to meet the principal needs of a breeding program within the scope of available resources. Varietal evaluation in the past was generally descriptive, designed primarily to facilitate the classification and identification of commercial varieties. Morphological characteristics made up the principal features of such systematic observation and recording. Plot yields were

sometimes recorded. Data on resistance to pests or to adverse environmental factors were based on natural, rather sporadic incidences. The evaluations were usually carried out by economic botanists, breeders, or agronomists.

Recent efforts at research centers such as the IRRI combine the characterization of cultivars and breeding lines by their morpho-agronomic features, the systematic determination of their reactions to major diseases and insect pests, and of their protein and lysine contents. Similarly, at the U. S. Department of Agriculture, considerable attention is given to the processing and cooking characteristics of foreign and domestic rices (Simpson *et al.*, 1965). Systematic characterization operations generally involve thousands of accessions. Such operations require the participation of researchers from several disciplines and often from different experiment stations.

A third type of systematic screening involves an empirical and intensive search for strains with a specific trait, when such a trait is urgently needed. The trait may be a high degree of resistance to a specific pathogen or insect, tolerance to an adverse climatic factor such as low air or water temperature, resistance to drought, or tolerance to a problem soil. Such screening usually involves hundreds or thousands of cultivars. The operations require ample technical information about the trait concerned, specialized equipment or facilities to identify different varietal types or to maximize varietal differences, and a mass-screening technique enabling the researcher to conduct the tests on a continuous basis and in a reproducible manner.

#### SIZE OF POPULATIONS FOR SCREENING

There are no hard and fast rules as to appropriate sample size of an accession for systematic screening. The researcher should consider both the inherent variousness of the trait concerned and the facilities and resources available to him. A small quantity of seeds may sometimes be a limiting factor.

As a general rule, fewer seedlings or plants are needed when the trait has high heritability, such as the reaction to a single bacterial blight isolate. Quantitative traits generally call for larger samples. The genetic composition of the accession may also determine the efficient sample size — a heterogeneous or heterozygous population requires a larger sample than does a pure or

homogeneous one (see discussion by Oka, 1969). When the researcher cannot effectively control the environment, he should choose an efficient experimental design and increase the number of replications of each accession. Discussions on sample size and sampling methods for research problems in irrigated rice are given by Gomez (1972).

#### METHODS OF SYSTEMATIC EVALUATION

Procedures and techniques for evaluating varieties vary with the expressivity of the trait concerned, the available magnitude of varietal differences, the level of varietal response being sought, the number of varieties to be tested, the desired degree of precision, and the manpower and physical facilities available. Moreover, methods for screening some traits are continually being developed or improved. Therefore, information is given only on the important traits. The references focus on testing. methods designed for screening large numbers of accessions.

#### **Agronomic traits**

Fertilizer response. The fertilizer response of a cultivar can be expressed either by the rate of increase in grain yield per unit measure of fertilizer or, in economic terms, the ratio of crop value from increased yield to the cost of fertilizer used. When tropical varieties differing greatly in growth duration are compared, it appears more meaningful to express the response as the rate of yield increase per unit of fertilizer per day.

Chandraratna (1961) has proposed the use of the quadratic equation,  $y = a + bx + cx^2$ , to evaluate variety × nitrogen interactions in place of the Mitscherlich curve. Also see IRRI Annual Report for 1973, p. 42-43 (IRRI, 1974).

For comparing and describing a variety of nitrogen response curves, the following parameters are useful (IRRI, 1974, p. 43).

- I)  $N_{max}$  nitrogen rate that maximizes yield
- 2)  $Y_{max}$  maximum yield
- 3)  $Y_I$  yield increase per unit of nitrogen applied based on  $N_{max}$  rate.

Heavy doses of fertilizers, especially nitrogen, tend to predispose cultivars to heavier infection by diseases or infestation by insects. The researcher can obtain additional information from fertilizer experiments by taking notes on the incidence of pest damages, followed by covariance analysis.

Resistance to lodging. Rice researchers use different ways and means to measure resistance to lodging. The commonly used criteria are Young's modulus (Hashimoto, 1963), bending moment (Seko, 1962), cLr or the lodging resistance factor (Chang, 1964, 1967), snap score (IRRI, 1965), breaking index of culm sections on the Salmon testing machine (Bollich, 1963), and resistance to pulling (Sharp, 1957). None of these methods is satisfactory, however, for use on a large number of test varieties.

When a large number of accessions are to be evaluated, the researcher may use the simple approach of gently pushing the culms of flowering plants back and forth at a distance of about 30 cm from the ground. This simple test indicates culm stiffness and resilience (Chang and Bardenas, 1965).

Observations made near and at maturity, of course, provide the best assessment of resistance to lodging. Notes should be taken on the growth stage at which lodging begins, the culm angle of lodged plants, and the percent of lodged plants in a plot. Additional notes could be taken on varietal characteristics such as sturdy vs. weak, and brittle vs. resilient (Chang and Bardenas, 1965).

Grain dormancy. Grain dormancy is determined from freshly harvested grains which have not been dried by heat. Germination tests of air-dried seed samples within a 2-week period following harvest in the wet season, or a 1-week period in the dry season, reveal varieties that have no dormancy. Successive germination tests of grain samples at intervals of 14 to 20, 35 to 50, and 80 days after harvest differentiate among weakly, moderately, and strongly dormant types (Chang and Tagumpay, 1973). The length of heat treatment at 50°C required to break dormancy indicates the intensity of dormancy in dormant varieties (Jennings and de Jesus, 1964).

Shattering. The ease of shattering has been tested by counting the percentage of shedded grains in a cloth sack during the process of transport and air-drying (Nagamatsu and Ishikawa, 1954), rolling a heavy cylinder over the panicles on an inclined board (Hanumantha Rao, 1935), or by mechanical devices (Ito, Inouye, and Tikai, 1968). But none of these techniques is suited for large-scale evaluation studies.

The simplest though rather crude method to ascertain ease of shattering is to grab a panicle at the hard dough stage with the

palm of the hand and apply a gentle rolling pressure. The percentage of shattered grains indicates whether the panicle is shattering (50 percent or more), intermediate (25-50 percent), or tight (few or no grains removed) (Chang and Bardenas, 1965).

# Physiological and ecological characteristics

Growth duration and photoperiod sensitivity. Planting test varieties at regular intervals to sample seasonal variation in daylength provides some information on photoperiod response at a given location. For more precise information, test the varieties under controlled photoperiods (see Vergara, Chang, and Lilis, 1972, for detailed discussion). With hybrid progenies or valuable material with few seeds, the separation of tillers from a plant allows a test of the plant to two or more photoperiods concurrently (Chang, Li, and Vergara, 1969). Types of varietal response to photoperiod are described by Chang and Vergara (1971).

*Growth analysis*. Techniques designed to estimate net photosynthetic production in plants can be found in the manual of Sestak, Catsky, and Jarvis (1971).

Air temperatures. Varietal response to low or high air temperatures can be assessed from different dates of planting, often repeated over seasons. More precise information can be obtained under controlled conditions in a growth chamber (IRRI, 1971, 1972, 1973).

Water temperature. The effect of water and soil temperatures on lowland rice can be evaluated from several dates of plantings in the field, or in tanks. Such experiments should be repeated over two or more years because of seasonal variation in temperatures. Growing potted plants in tanks with circulating water at specified temperatures provides more reliable results (Ormrod and Buntner, 1961; Adair, 1968; IRRI, 1972).

Deep water. Varietal tolerance to deep water can be determined by planting the test varieties in fields where flooding is known to occur every year. Stations designed to make use of natural floods have been set up at Huntra in Thailand and at Habiganj in Bangladesh. To obtain varietal reactions to controlled water depths, however, large tanks or ponds that can accommodate hundreds of lines have proved to be most useful (Yantasast, Prechachart, and Jackson, 1970; Jackson et al., 1972).

Submergence. Temporary submergence of plants by flash

floods may occur in areas with normally shallow water. Varietal tolerance to submergence is best determined from test plants submerged in ponds or large tanks (IRRI, 1972).

Drought. Techniques for evaluating resistance to drought are being developed or improved. Field testing in the wet season is best carried out by planting on two or more dates at several sites, hoping to sample dry spells at different stages of growth. By planting a large number of single-row plots in the dry season, with controlled irrigation, marked varietal differences in drought resistance have been observed. These differences are reproducible and appear to be related to reductions in yield following drought (Chang, Loresto, and Tagumpay, 1974).

The varietal differences thus obtained are probably indications of drought avoidance expressed in terms of root development and stomatal resistance to water loss. Varietal tolerance to drought and to heat needs to be assessed by more refined techniques (IRRI, 1973, 1974, 1975).

Recovery from desiccation or extreme water stress is determined from pot or tank experiments where soil moisture is monitored (IRRI, [1964], 1971; Chang, Loresto, and Tagumpay, 1972; IRRI, 1974).

Strong winds. Damage to rice leaves by strong winds can be assessed by the extent of leaf blades torn to shreds or horizontally broken and the percentage of leaves so affected (IRRI, [1967]; Alluri, Vergara, and Visperas, 1973).

# Adverse soils and nutritional disorders

An array of nutritional disorders or physiological disturbances have been identified when rice is grown on wide ranges of soils in either anaerobic (flooded) or aerobic (upland) conditions. Differences in varietal response to deficiency or excess of an element are indicated by a range of plant symptoms and differences in growth rate and yield, when test varieties are grown in such soils either in pots or in the field. Techniques for determining varietal responses to adverse soil factors have been described by Ponnamperuma and Castro (1972) and in IRRI annual reports (1973, 1974). Discussions on nutritional disorders and laboratory techniques for analyzing such disturbances were provided by Tanaka and Yoshida (1970) and Yoshida *et al.* (1972).

#### Diseases

Blast. Testing rice seedlings for reaction to the blast fungus (Pyricularia oryzae) can be done in the field or in the

greenhouse. The procedure for setting up a blast disease nursery of uniform design in the field has been described by Ou (1965). Neck blast can be induced by inoculating the boots with the fungal culture (Ou and Nuque, 1963). Greenhouse tests are usually inoculated by spraying the seedlings with a conidial suspension (Anderson and Henry, 1946) or by injecting the spore suspension into the leaf-sheaths (Kurigayashi and Terazawa, 1953). Cut sections of leaf sheath may also be used for inoculation (Sakamoto, 1949).

Bacterial leaf blight. The pathogen (*Xanthomonas oryzae*) can be inoculated by pricking the leaf tissues with pins (Mizukami, 1966; IRRI, 1966), dipping the seedlings in a bacterial suspension (Mizukami, 1966), or cutting leaf tips with scissors dipped in the bacterial suspension (Kauffman *et al.*, 1973).

Bacterial leaf streak. Cultures of Xanthomonas translucens f. sp. oryzicola are inoculated by spraying 3-week-old seedlings with a bacterial suspension and incubating the plants in a high humidity chamber for 15 hours (IRRI, 1968).

Sheath blight. This fungus disease caused by Thanatephorus cucumeris (formerly Corticium sasakii) can be tested in a variety of ways. Plants grown in flooded fields can be inoculated by placing diseased tissues around the base of plants or inside the clusters of tillers and wrapping the plants with paper to keep the culms moist. An upland nursery similar to that for blast has been used with success at IRRI. Seedlings are inoculated at 3 weeks after seeding with a fungal culture reared on rice straw (see Ou, 1972).

Tungro and grassy stunt. Mass screening for varietal reaction to the two major virus diseases of the tropics, tungro and grassy stunt, has been developed and recently improved (Ling, 1969; Ling, Aguiero, and Lee, 1970; IRRI, 1973, 1974). A caging technique using individual plants for testing was used by Indian workers (Shastry, John, and Seshu, 1972). Field screening of large numbers of varieties and lines is also feasible, but the uniformity and severity of infection are affected by environmental conditions (Ou, 1972; IRRI, 1974).

Methods for determining varietal reactions to other rice diseases were discussed by Ou (1972) and Toriyama (1972).

### **Insect pests**

*Stem borers*. Mass screening for resistance to stem borers has been discussed by Pathak *et al.* (1971) and Pathak (1972).

Leafhoppers and planthoppers. Testing rice seedlings for susceptibility to damage by the leafhoppers (Nephotettix virescens and N. nigropictus) and the brown planthopper (Nilaparvata lugens) in a screenhouse has been described by Pathak, Cheng, and Fortuno (1969) and Pathak (1972). The life span of insects feeding on individually caged plants provides another criterion for determining varietal reaction to this group of sucking insects.

Gall midge. Mass screening for the rice gall midge (*Pachydiplosis oryzae*) is usually performed in the field at sites of naturally heavy infestation (Ventakaswamy, 1969; Pongprasert *et al.*, 1972; Shastry *et al.*, 1972).

*Stem maggot.* Reaction to the rice stem maggot (*Chlorops oryzae*) can be screened in the field, and with controlled infestation in a greenhouse (see Pathak, 1972).

### Physical characteristics of the grain

*Size and shape.* The dimensions of the rice grain can be readily determined by measuring well-developed grains on a small ruler. The image produced on a calibrated plate by rice grains, with a photo-enlarger as the light source, makes possible the precise determination of grain dimensions.

FAO has recently revised its classification scheme for grain size and shape. The FAO standards (FAO, 1972) along with those used by USDA rice workers (Adair *et al.*, 1973) are given below

Size (length)	USDA scale for brown rice	FAO scale for milled rice					
Extra long	More than 7.50 mm	7.00 mm and above					
Long	6.61-7.50 mm	6.00-6.99 mm					
Medium	5.51-6.60 mm	5.00-5.99 mm					
Short	Less than 5.51 mm	Less than 5.00 mm					
Shape (length/ width ratio)	USDA scale for brown rice	FA0 scale for milled rice					
Slender	More than 3.0	More than 3.0					
Medium	2.1-3.0						
Bold	Less than 2.1	2.0-3.0					
Round	_	Less than 2.0					

Milling quality. The proportions of brown rice, milled rice, and head (whole-kernel) rice to grain (rough rice) are expressed

on a percentage basis by weight. These are called brown rice yield (or hull percentage), total milled rice yield, and head rice yield, respectively.

Endosperm characteristics. Translucency, the presence and size of chalkiness (white belly, white center, and white back), and the condition of the "eye" (degermed end) are rated at IRRI on a coded scale of from 0 to 5. A lower number indicates a more desirable class (IRRI, 1966, p. 84-85).

Other physical evaluation tests are described by Adair *et al.* (1973). The test-tube miller facilitates the milling of large numbers of small samples of brown rice (Khan and Wikramana-yake, 1971).

# Nutritive and cooking quality

*Protein and lysine contents.* A technique for large-scale screening of rice cultivars for protein content has been developed by Juliano *et al.* (1968). Lysine content can be readily determined by the method of Juliano, Antonio, and Esmama (1973).

Cooking characteristics. The determination of amylose content has been simplified by Juliano (1971). An estimate of gelatinization temperature by the alkali-digestion method has been in wide use since it was developed by Little, Hilder, and Dawson (1958). Elongation ratios of milled rice can be obtained during the soaking and cooking processes (IRRI, 1971). Recently a gel consistency test has been devised to critically evaluate eating quality (Cagampang, Perez, and Juliano, 1973).

Other quality tests have been described by Simpson *et al.* (1965) and by Juliano (1972).

#### FIELD VERSUS LABORATORY TESTING

Laboratory tests are generally designed to facilitate rapid determinations where large number of accessions and a specific causative factor are involved. Tests in the laboratory or glasshouse or nursery beds are usually made on seedlings. They are specially suited for determining nutritional disorders, diseases or insects that appear in the seedling or juvenile stage, and phytotoxicity ratings.

However, laboratory tests cannot entirely replace field plantings, which cover the entire life cycle of the plants. Field tests not only allow large number of accessions to be tested and cost less for physical facilities, they also offer the advantages of 1) sampling environment  $\times$  causative factor interactions, 2) sampling variations in the biotic factor (e.g. pests), 3) tracing dynamic changes related to the growth of plants and continuous changes in the climatic and edaphic environments, and 4) providing more reliable information on grain yield and quality by allowing plants to grow to maturity in replicated plots. Results from laboratory tests generally require verification under field trials in the same manner that quick tests for soil fertility are generally confirmed by plot trials.

#### METHODOLOGY OF EVALUATION

In the evaluation process, the researcher should recognize that a seed sample of a cultivar is not necessarily pure and that it usually contains more than one strain with respect to the trait being tested for. Similarly, a given disease or insect cannot be adequately represented by one isolate or sample. Genetic diversity within any biological entity, whether cultivar or pest, is a universal phenomenon.

In testing varieties for reactions to pests, the inclusion of several representative strains or biotypes of a pest enhances the scope of screening. Moreover, the researcher should know the specific relationship between genes in the host plant for resistance (or susceptibility) and the genes in the pathogen for virulence (or avirulence). It is necessary, for useful results, to test large numbers of accessions and to include representative strains of a pest in the search for genetic resistance to it (see Chang *et al.*, 1975).

It is also essential to include an adequately large sample of an accession in the evaluation process if the desired phenotype is to be found in the population, particularly when the population contains different genotypes or when the expressitivity of the trait is rather low, or both. When a researcher is dealing with quantitative traits such as protein content and insect incidence, both the sample size and sampling method should be carefully studied and chosen. Oka (1969, 1975) has discussed the relationship among the fraction of genetic information desired, population size, and genetic variability within a population as well as between populations. Suggested sample size and sampling method for stem borer incidence, protein content, and other quantitative traits of rice are provided by Gomez (1972). The

coefficients of variation for many plant and grain characteristics found in Japanese cultivars are given by Ito (1965, p. 207).

The researcher should know the specific environmental conditions under which the trait being evaluated would be expressed to the maximum extent. In many cases the researcher may have to choose between the most favorable conditions for the manifestation of a trait and the most practical conditions under which the expression can be readily and inexpensively reproduced from experiment to experiment. In most instances, measures for controlling environmental conditions are necessary.

The inclusion of one or more "control varieties" of known reaction to a biotic or ecological factor is an absolute necessity in all screening work. In field tests principal cultivars of current importance should be included as controls.

When a researcher has identified a promising source for a desired trait, he should not be satisfied with that one source only. Since genetic diversity is needed in future varieties, the worker should look for additional sources of such a controlling gene or genes. To identify genetically diverse sources, the researcher should compare a new source with the known source by genetic experiments. Examples of such tests for allelism have been reported for semidwarfism (Chang and Vergara, 1972) and for resistance to the leafhoppers and planthoppers (Athwal and Pathak, 1972). Such a process of genetic cataloging would help rice breeders choose distinct sources of desired genes from a large number of accessions which are often duplicate carriers of a smail number of genes.

An efficient evaluation program calls for certain amounts of fundamental research on the subject as well as interdisciplinary collaboration. Examples of such a multidisciplinary approach in searching for resistance to diseases and insect pests are discussed by Chang *et al.* (1975).

When one or more promising sources of useful traits are found in samples coming from one or more areas of known genetic diversity, the national center should send collection teams into those areas to collect more intensively for the desired types, as well as the wild relatives. The collectors could also expect to find a diversity of pathogenic or insect biotypes in areas of large varietal diversity. This is one way that findings from evaluation can help in further enriching the gene-pools of the national collection.

#### DISTRIBUTION OF SELECTED MATERIAL FOR UTILIZATION

Strains of potential value should be widely tested and utilized in breeding or other research programs. Testing by researchers at different sites will indicate the stability of a trait and may reveal genotype × multi-environment interactions which can have specific applications. Some of the accelerated progress in varietal improvement has resulted from concurrent collaborative testing at several sites. The identification of outstanding blast-resistant varieties was made possible by international cooperative testing at more than 60 sites.

To facilitate regional collaborative testing, the seedstocks of promising varieties or strains should be made available to workers at other sites. Information on promising materials warrants dissemination through newsletters, journals, scientific conferences, or special circulars. Alternatively, the strains can be entered in international testing programs such as the international variety nurseries, international blast nurseries, or the bacterial leaf blight nurseries being coordinated by the IRRI staff (IRRI, 1974).

Sufficient seedstocks of promising accessions should be raised for distribution to other workers who ask for them. Strains which were selected from an apparently heterogeneous variety should be so designated as to distinguish them from the original accession.

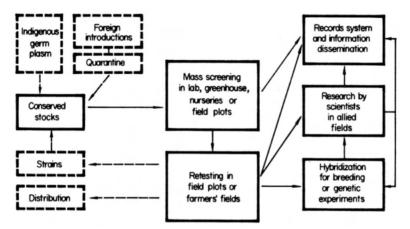
Promising findings obtained at different experiment stations or colleges should be transmitted to the international center so that the scope of utilization by other institutions can be expanded within a short time.

A generalized procedure designed for evaluating and utilizing rice germ plasm is shown in Fig. 4.

The interdisciplinary rice improvement effort of IRRI scientists under the Genetic Evaluation and Utilization (GEU) Program is described in "Research Highlights for 1973" (IRRI, 1974a) and the IRRI Reporter issue 2/74 (IRRI, 1974b).

#### GENETIC ASPECTS OF UTILIZATION

Since wide adaptiveness and high yield potential have been largely achieved by the development of the semidwarfs, those genes that many rice breeders are searching for are primarily those needed to eliminate defective features such as susceptibility



4. Flow chart outlining the necessary steps in the evaluation and utilization of rice germ plasm in relation to conservation and dissemination. Operations indicated by solid lines and arrows are directly related to evaluation, utilization, and information storage and retrieval. Dotted lines and arrows indicate indirect activities.

to certain diseases and insects, or a lack of specific adaptation to adverse physical or edaphic environments. To raise the physiological efficiency of the crop is another long-range goal.

Although a considerable number of resistant sources have been found for a major disease or insect, in each instance only a few non-allelic genes are available to rice breeders (Chang *et al.*, 1975).

In removing susceptibility to a specific disease for example, the breeder is inclined to use a highly resistant source regardless of other agronomic traits involved. The process generally aims to transfer a small segment of chromosome from the resistant parent. Experience at IRRI had indicated that the probability of obtaining more desirable progenies is higher when the resistant parent has fairly good agronomic features rather than very poor features. Breeding efforts at several national centers have shown that programs specially designed to correct one major trait such as susceptibility to blast or to lodging have not produced agronomically promising varieties.

To transfer resistance to a pest into an adapted type, the breeder tends to choose the highest level of resistance available. He then has to choose between resistance to a few races controlled by one or a few major genes (oligogenic or vertical type) and resistance to the entire population of the pest governed by many genes (polygenic or horizontal type). The major-gene resis-

tance to a specific race is easy to transfer and gives more spectacular effects when first introduced. But this type of resistance generally breaks down rapidly after the resistant variety is grown commercially. Japanese breeders have experienced the frequent breakdown of blast resistance in new hybrid varieties when resistance is controlled by one gene. Continuous rearing of brown planthoppers on the resistant variety Mudgo in screened cages at IRRI has resulted in the appearance of a new biotype of planthoppers that can attack Mudgo in spite of the resistance gene *Bph*-1 present in the variety.

On the other hand, true horizontal resistance is difficult to find. Resistance that is non-specific to different races has been found in other crops but is yet to be identified in rice. A more practical approach is to incorporate into adapted types many genes that will confer resistance to several races. The combined effect of different genes will provide a reasonable level of stability in host resistance.

Lying somewhere between the major genes and the polygenes is a group of polymeric genes, with cumulative and unequal effects, that often operate collectively to produce a range of variation in a trait. Such anisomeric genes have been shown to control the basic vegetative phase of growth duration or the strength of grain dormancy. We might expect this kind of gene interaction in controlling a component of the resistance to an adverse factor, physical or biotic.

When an accession lacks a desirable feature, it is not always true that the accession lacks the desirable alleles. In rice, several cases have been found where a suppressor (inhibitor) gene blocks the expression of the desired gene when both are present in one genotype. Such gene interactions have been found for short plant height, anthocyanin pigmentation, photoperiod sensitivity, insect resistance, and disease resistance. Therefore, a breeder may expect to find resistant progenies from susceptible × susceptible cross combinations, if he has made many crosses involving diverse parents and is testing large segregating populations with the appropriate technique.

In addition to multiple genes of resistance to a disease or an insect, resistance to multiple diseases and insects should be the goal of a breeding program. For virus diseases, resistance to both the virus and the insect vector is necessary to provide adequate protection. By systematic screening, TKM-6 is now

known to have resistance to bacterial leaf blight, the striped borer, and the green leafhopper. Similarly, DV-29 is resistant to tungro, green leafhopper, and brown planthopper. Such varieties offer distinct advantages for use as parents.

A strain or a variety can be repeatedly tested and selected to attain a very high level of phenotypic purity. Residual variousness in minute features (polymorphism) will be revealed if refined tests, such as isozyme analysis and inoculation with specific pathogenic isolates, are applied. The persistence of genetic heterogeneity in spite of rigorous pedigreed selection indicates a potential in the retention of genetic variousness that might be used in the future to meet needs for long-range adaptation.

While the current interest in the wild species primarily concerns their resistance to pests, the wild forms offer potential in adapting themselves to changing adverse environments because of their inherent heterozygosity. Such genetic plasticity has enabled the wild forms to survive marked changes in their physical and biotic environments. Thus, the wild forms offer a broad genetic base for long-range adaptation.

#### DATA RECORDING, PROCESSING, AND RETRIEVAL

Before the collection, characterization, and evaluation operations grow in size and scope, the national center should plan and establish a system for recording and processing data on the genetic resources. The system should be so designed that it can be linked to an international seed bank or data bank for the crop. The records system should include information on the conserved stocks, systematic description of morpho-agronomic traits, results of evaluation, and maintenance and distribution records. A uniform system of recording will help in collating and retrieving information originating at different national centers.

Since 1965 the FAO/IAEA Working Group on International Standardization in Crop Research Data Recording has worked toward the development of guiding principles in uniform documentation (see Konzak and Dietz, 1969). IRRI technical bulletin 4 on morphology and varietal characteristics (Chang and Bardenas, 1965) represents the first effort in cereals to devise a uniform recording system for a crop species. Additional suggestions on the growth stages of rice along with the morpho-

agronomic coding system were adopted with success in the FAO/IAEA-sponsored uniform yield trials for promising mutants and hybrids which were conducted during 1966-68 at 13 locations in 10 countries of Asia, Europe, and South America (Chang, 1970). The morpho-agronomic coding system was used in preparing the IRRI variety catalog (IRRI, 1970) which included 8,628 accessions and 40 items each. The accession number, variety name, origin, seed source, and 40 items were entered on two IBM punch cards and are amenable to machine sorting. While computer processing is not an absolute necessity for all centers, large-volume processing requires a computerized system.

For a national center, complete records on past collections must be maintained. The record form for field collectors of rice and the registration operations were described by Chang *et al.* (1972). For genetic resources centers involved in extensive field collection activities of different crops, record forms suggested by Rogers, Snoad, and Seidewitz (1975) offer additional merit in data processing and retrieval.

At most national centers, documentation should begin at the registration phase. Basic items to be systematically recorded are crop species code, accession number, origin, seed source, accession name, the international registration number (or accession number), variety-group, morpho-agronomic data, evaluated results, and seed stock number (or numbers) along with the crop season code. When space is available on the data card, information on duplicate seed samples deposited at a seed bank, seed distribution, year and seed increase, and seed quantity, should also be entered. A record of stocks maintained at different state or provincial centers will be helpful to the national center in overall planning for seed increase, distribution, and maintenance.

The data form used at IRRI for recording morpho-agronomic traits in the field is shown in Appendix III. Additional data are obtained from laboratory measurements of grain features and various evaluation tests. These data along with information recorded at the registration phase are coded, punched on cards, arranged in alphabetical and numerical order, and printed in the catalog form as shown in Appendix I. It can be modified to suit the varying needs of different national centers.

Every evaluation operation is an experiment. The record system should include an identifying card for each variety or selection entered in the various tests for the master record file, an experimental plan describing the treatments and varieties and experimental design, notebooks, and a set of data records (one record for each plot) for complex experiments. Observations are recorded in the notebooks or on the printed cards. Data are transferred from the notebook or a voice-recorded tape to the data cards for statistical analysis. When a computer is used to process the data, summary tables can be machine printed. The notebooks and statistical data are returned to the researchers concerned for interpretation, preparation of reports, and utilization. Data of different experiments entered on punch cards or magnetic tape are much easier to retrieve and to collate than handwritten data entered in several notebooks, especially when the testing covers several years.

A uniform system for recording and processing cereal research data has been proposed by McNeal *et al.* (1971). An example of the experimental plan and field book excerpted from the FAO/IAEA mutant trials and the international rice yield nursery, both of which are coordinated by the IRRI, is shown in Appendix IV. A standard scoring system for rice experiments (IRRI, 1975) has been recently prepared and adopted by rice researchers who participate in the international rice testing program.

Computer programs are generally written in the FORTRAN computer language. Recently a TAXIR program (for taxonomic information retrieval) has been proposed to facilitate the storage and retrieval of data on accessions at seed or data banks (Hersh and Rogers, 1975). Such a program would permit a bank to find accessions which, for instance, will combine 100-day maturity, cool tolerance, blast resistance, stem borer resistance, slender grains, intermediate amylose content, and low gelatinization temperature. The adoption of a uniform recording system is a prerequisite to an efficient information retrieval system.

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# **Genetic Conservation**

#### LONG-RANGE CONSERVATION

Long-range conservation aims to preserve the genetic totality of populations in such a way that the loss of certain genotypes in the population or a marked change in the gene frequency of many loci will be minimized. In other words, the population structure of the original accession needs to be so maintained that no particular genotype or genotypes are being selected for at the expense of losing others in the process of rejuvenation.

In self-fertilized species such as *O. sativa*, long-range conservation is most conveniently accomplished by long-term seed storage and a minimum number of cycles of seed rejuvenation. This method requires careful handling in seeding and harvesting, roguing, minimization of cross-pollination, and seed storage in vacuum at low temperature. If facilities are not available for storing seed in vacuum at low temperature, viability can be retained for several seasons by drying the seed to 9 percent or lower moisture content and storing in airtight containers (see medium-term storage conditions described by Chang *et al.*, 1972).

Planting should be made at sites where no physical or biotic environment will select against unadapted or susceptible genotypes. Long-term seed storage minimizes hazards of introducing genetic changes in a population during rejuvenation. However, a sufficiently large sample needs to be stored in order to guard against changes in the genetic composition of the sample due to differential viability among seeds. On the other hand, such a maintenance program is rather inefficient when one compares the large number of accessions being maintained to the rather small number of useful genes being included.

For wild taxa which are prone to cross-pollinate, bagging of panicles is necessary to prevent outcrossing and to cope with grain shedding. In perennial forms, vegetative propagation is an efficient way of preserving a heterozygous strain, provided that the plants are protected from virus diseases and insect pests.

For heterogeneous populations such as samples of *O. rufi- pogon*, *O. nivara* and the 'spontanea' rices, a population often contains a number of discernible genotypes that differ in morphological and physiological aspects. To preserve the population structure, the individual strains should be separately maintained as sub-populations and later a sample each from the sub-populations should be taken to reconstitute the population. For many of the wild-growing taxa that have specific growth conditions, preserving the populations in their natural habitats offers distinct advantages. However, such natural reserves need to be protected by national governments.

An alternate approach to conserve genetic variability is to composite a selected group of varieties and perpetuate the mixture without human selection. Another method is to composite a large number of  $F_1$  hybrids or the progenies and allow the bulk population to undergo some degree of cross-pollination and natural selection. Both methods present technical and operational problems, however. Another possibly useful method is storage of cells or tissues of different genotypes in a synthetic medium under freezing conditions. Research is needed to adapt these techniques for use on rice. Some of these techniques will be available only to well-equipped national and international centers.

#### Long-term seed storage

*Need for different types of storage.* After an accession has completed the seed multiplication and initial description (or evaluation) process, the seed stock should be divided into several lots. One portion should be set aside for evaluation studies in the next few years. Methods designed for short-term storage (2 to 3 years) will be adequate for this seed lot (see Chang *et al.*, 1972).

A second portion should be placed in a vault designed for medium-term storage (5 to 10 years or slightly longer). If a state or provincial experiment station does not have such facilities, a seed lot should be sent to the national center for storage and for further evaluation.

Seed stocks for rejuvenation should be separately stored from those for distribution.

The national center should deposit a seed sample (15 g or more) at an international seed bank for preservation. Preservation requires facilities for long-term storage.

Preparing seed for long-term storage. Seed intended for long-term storage should be harvested as soon as the seed is physiologically mature — the optimum time is when about 80 percent of the spikelets have reached the hard dough stage. Delayed harvesting exposes seeds to damage by insects or fungi or adverse climate. In threshing, damage by high-speed threshing should be avoided. Clean seeds thoroughly before drying, fumigation, and storage. Complete the operations of harvesting, transport, threshing, and cleaning within the shortest period possible.

Drying by artificial heat should first be made at a rather low (40°C) level when the moisture content exceeds 18 percent. The temperature can be raised to 50°C when the moisture content is below 14 percent. Slow low-temperature heating in a dry atmosphere is better than fast high-temperature treatment in humid air. In the tropics, the last phase of seed drying should be carried out in a dehumidified (airtight and air-conditioned) room and in a heating cabinet containing desiccants so that the moisture content can be lowered to 8 percent or less without using temperatures exceeding 50°C.

Two rules of thumb may indicate the relationship between storage conditions and seed moisture content (Harrington, 1972): "life of seed is halved by each one-percent increase in seed moisture when moisture content is between 5 and 14 percent, and the life of seed is halved by each 5-degree increase in seed temperature, when the range is between 0° and 50°C. These two rules apply independently." Therefore, long-term seed storage requires low temperature, low relative humidity, low volume of air around seeds, and low oxygen content.

If seeds are stored at moisture contents above 8 percent and ambient temperature above 5°C, fumigating the seeds before storage is necessary to control insects. A mixture of carbon tetrachloride and ethylene dichloride appears to be the most convenient chemical treatment (Chang, 1974). Fumigation is unnecessary for seeds to be stored near or below freezing temperatures.

Conditions for medium- and long-term storage. The storage room should be insulated and airtight, preferably located on the north side of a permanent building. Refrigerate the room to an

economically feasible temperature (15°C or below). Our experience has shown that it is highly desirable to have the cold storage room connected with the seed laboratory or seed processing area by an air-conditioned room where final seed drying, sorting, and packing can be performed. This area will serve as a buffer zone between the cold room and the unrefrigerated area and makes it easier to maintain the low temperature and low humidity in the storage room.

Since refrigeration alone cannot lower the relative humidity effectively below 15°C, it is essential to place dried seeds in moisture-proof containers. Aluminum foil-polyethylene envelopes are excellent for storing small seed samples. A desiccant such as silica gel placed inside a large, sealed container further reduces the moisture content of seed. Fill the container completely with seed or seed samples so that little air remains. Large glass jars of 2-gallon capacity used at IRRI makes it possible to check on the dryness of seed by a glance at the color of indicating silica gel inside the jars (Chang, 1972).

For working collections, the storage conditions at IRRI (8–9 percent seed moisture, 3-4°C, 60–75 percent relative humidity) have proved to be effective and practical for medium-term storage. Ten-year-old seeds of tropical varieties have more than 90 percent viability. The viability of japonica varieties, however, drops at a faster rate. For long-term storage, the use of vacuum cans and below-freezing temperatures gives closer to ideal conditions. Only the National Seed Storage Laboratory in Japan has such storage conditions.

It is difficult to predict seed longevity under given storage conditions. Roberts (1975) has proposed a formula to make such predictions, though the biological nature of seeds often defies strictly physical considerations. It is necessary to check periodically on the viability of stored samples in order to plan for rejuvenation operations. At IRRI germination tests are made twice a year on three control varieties stored in quantities larger than those of the conserved stocks.

Storage at duplicate sites. To insure safekeeping, seed samples should be stored at two or more seed banks. IRRI stores a duplicate set of seed samples at the U.S. National Seed Laboratory, Ft. Collins, Colorado.

Operating a seed storage laboratory. A storage lab must have competent personnel to continually run the technical operations

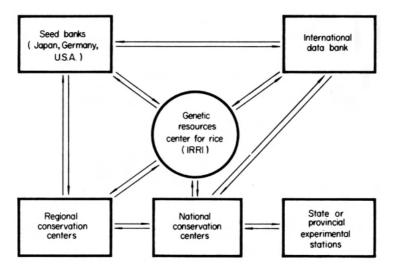
and to maintain the physical facilities. Complete records should be kept on the source, quantity, age, viability, and distribution of seeds. Such records should be brought up to date at regular intervals.

#### International collaboration in genetic conservation

Operations related to the long-range genetic conservation of a staple food crop such as rice are beyond the capability of a single institution. Although the IRRI has taken on the global responsibility of conserving rice germ plasm, we find it necessary to enlist the collaboration of other international and national organizations in achieving the goals. Since 1971 Bangladesh, Burma, Indonesia, Khmer, Malaysia, Nepal, Sri Lanka, and South Vietnam have collaborated with IRRI in carrying out field collections for minor varieties and specialty types. Thousands of samples have been assembled from many remote areas. The collection activities in tropical Asia have progressed so well that we can expect to conserve a substantial segment of the rice germ plasm during this decade. Collection of rices in tropical Africa needs to be intensified, however.

The evaluation phase will exceed the collection phase in scope and depth. International and inter-institutional collaboration are indispensable to evaluate the collected materials. The recently initiated International Rice Testing Program serves as the channel for worldwide evaluation (IRRI, 1975).

The preservation of rice germ plasm will require a greater degree of international collaboration than in the past. The capabilities of a crop-specific institute such as IRRI in genetic conservation have been described (Chang et al., 1975). But one institution at one site cannot adequately manage the conservation of diverse germ plasm without the danger of losing unadapted or susceptible stocks in the process of seed increase or renewal (Chang, 1972). While IRRI will continue to provide rice researchers with seed samples of the entire world collection, a network of institutions is needed to share the various responsibilities related to long-range preservation and seed distribution (see Frankel, 1975). Ideally the system should consist of a genetic resources center (IRRI), two or more seed banks (U.S. National Seed Laboratory, National Seed Storage Laboratory of Japan, etc.), several regional conservation centers, and all of the national conservation centers concerned.



5. Proposed network of international and inter-organizational collaboration for the seed increase, seed and data storage, distribution of seed and related information, and rejuvenation phases of genetic conservation of rice.

Long-term seed storage of the world's entire collection should be handled by the genetic resources center at IRRI with the assistance of the seed banks. Each of the regional conservation centers should maintain by seed rejuvenation the germ plasm truly indigenous to the geographic region concerned and supply viable seed stocks for storage at IRRI and the seed banks as well as stocks (that cannot be readily multiplied at IRRI) for evaluation and distribution. We propose that the national centers of China and that of Japan collaborate with IRRI in rejuvenating the temperate-zone varieties of Asia, that the U.S. Department of Agriculture and the Centro Internacional de Agricultura Tropical look after the stocks of the Americas, that the International Institute of Tropical Agriculture, the West Africa Rice Development Association and other collaborating institutions in Africa maintain the African rices (O. glaberrima), and that the IRRI rejuvenate the tropical rices of Asia. Each national center should maintain working collections of national interest and conserve natural reserves for indigenous wild forms. The genetic resources center will coordinate the various operations related to seed exchange and information retrieval.

The working relationship among the proposed institutions is illustrated in Fig. 5. We hope that rice researchers interested in

genetic conservation support and join this scheme of collaboration.

The training of competent personnel to execute operations related to characterization, evaluation, and maintenance is an area deserving immediate attention. The national conservation centers are in a position to identify promising workers to receive training at an international genetic resources center or universities offering similar programs.

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**6.** Inside the medium-term seed storage room at IRRI – seeds are stored in 2-gallon jars containing silica gel;  $3-4^{\circ}$ C, 60-70% R.H.

# **Appendixes**

A-

Appendix 1. A working key to the valid species of *Oryza* (modified after Roschevicz, 1931; Chatterjee, 1948; Tateoka, 1963; Bardenas and Chang, 1966; Tateoka. 1965; Sharma and Shastry, 1965).

-1 Sterile lemmas present
B-1 Sterile lemmas linear or linear lanceolate
C-1 Ligule of lower leaves 14-45 mm. long, tips acute
D-1 Annual; leaf blades narrow; no rhizomes; spikelets persistent,
4-8.5 mm. long, 2-4 mm. wide; cultivated; diploid sativa
D-2 Annual; semi-erect to decumbent growth; without rhizomes; basal
internodes spongy; spikelets oblong and deciduous; short anthers;
Asian and Oceanian origin; diploid nivara
E-1 Perennial; erect habit; branched, spreading rhizomes; elliptic
pollen; African origin; diploid longistaminate
E-2 Perennial; prostrate or floating habit; weak rhizomes; long anthers; adventitious roots and extra-vaginal branching at highe nodes; long internodes; lax panicles; long, slender spikelets Asian origin; diploid rufipogon (Asian race
E-3 Perennial; semi-erect habit; stoloniferous culms; long awns and
long spikelets; American origin; diploid rufipogon
(American race)
C-2 Ligule of lower leaves shorter than 6 mm., tip round or truncate
D-3 Sterile lemmas almost equal in length and similar in structure to
lemma and palea; ligule with a fringe of hairs at the apex; leaves
broad, American origin, perennial; tetraploid grandiglumis
D-4 Sterile lemmas considerably shorter than the lemma and palea
E-4 Annual; plants erect; leaves glabrous to slightly scabrid; panicles
more or less compact; lemma and palea perfectly or almost
perfectly glabrous; sometimes hispid; spikelets usually awnless or
short-awned; main panicle axis without secondary or tertiary
branches; spikelet length between 7-9 mm.; spikelet width
2.9-3.6 mm.; tip of sterile lemmas acute; African origin;
cultivated; diploid glaberrime
E-5 Annual; plants erect to spreading; panicles open, main axis with
secondary or rarely tertiary branches; lemma and palea hispid
spikelets 7.8–11.0 mm. long, 2.8–3.4 mm. wide; always awned (10 cm. or longer); awns bristled; sterile lemmas 2.1–5.0 mm.
long, acuminate at tip; African origin; diploid borthii <sup>3</sup>
iong, acummate at up, Amean origin, uipioid boring

<sup>\*</sup>Formerly known as O. breviligulata; includes the weed race 'O. stapfii Roschev.'

	Perennial; main axis of panicle slightly woolly pubescent at the base of primary branches, the rest smooth and glabrous; axis increasingly hispid-scabrous toward the tip; awns less than 5 cm. long; sterile lemmas linear or linear lanceolate; rhizomatous; Australian origin; diploid
	more than 7 mm. long; American origin;
F-8	tetraploid
L-0	than 2 cm broad
	F-2 Width of spikelets less than 2 mm.
	G-3 Panicle branches not spreading, length of
	spikelets 4.5-6.0 mm.; width 1.5-2.0 mm.,
	culm base slender, hard and not spongy; ligule
	less than 3.5 mm.; never split, hard, flexuous
	with fine bristles; sterile lemmas acuminate and
	narrowly triangular; African origin; perennial;
	mostly diploid, few tetraploid eichingeri
	G-4 Length of spikelets 3.7-4.7 mm.; width usually
	less than 2 mm.; panicle small with spreading
	branches; awned (2 cm. or less) or awnless;
	Asian origin; perennial; tetraploid minuta
	F-3 Width of spikelet more than 2 mm.
	G-5 Length of spikelets more than 5 mm.; width 2.0-2.5 mm.; culm base soft and spongy, ligule longer than 3.0 mm.; soft, and split when
	dried; straight or flexuous with rigid bristles;
	panicle loose with spreading branches; sterile
	lemmas acute and triangular; African origin;
	perennial; tetraploid or diploid punctata
E-9	Ligule glabrous or hairy; length of spikelets less than
	5 mm.; width almost always more than 2 mm.;
	awns often shorter than 2 cm., or awnless;
	occasionally rhizomatous; Asian origin; perennial;
	diploid officinalis
A-2 Sterile lemmas sub	
	erile lemma and palea granulate; sterile lemmas minute,
	ered from base; usually awnless
_	oblong to elliptic oblong, shorter than 7 mm.; perennial;
aipioia	granuluta

C-4 Spikelets narrowly oblong to lanceolate, longer than 7 mm.;

perennial; diploid meyeriana
B-3 Surface of lemma and palea not granulate; awned; spikelets 8-17 mm.
long
C-5 Lemma ciliate along keel, without wing; leaves herbaceous
D-5 Awns 6-15 mm. long; sterile lemmas shorter than lemma; perennial; tetraploid <i>ridleyi</i>
D-6 Awns 16-36 mm. long; sterile lemmas as long or longer than lemma; perennial; tetraploid longiglumis
B-4 Surface of lemma almost smooth with fine longitudinally dotted stippled surface
C-6 Spikelets linear, 1-2 mm. wide; awns 6-17 mm. long; sterile lemmas always much shorter than lemma; annual; diploid
A-3 Spikelets 1.5-1.75 mm. long schlechteri
NOTES:  a. Excluded from the key are taxa of doubtful validity or names of uncertain application:  O. abromeitiana, O. collina, O. fatua. O. glumaepetula, O. malampuzhaensis, O. perennis,  O. Schweinfurthiana. O. stapfii and O. ubanghensis.  b. Removed from the genus Oryza are taxa formerly known as 'O. angustifolia', 'O. perrieri' and 'O. tisseranti' - to the genus Leersia; 'O. coarctata' (renamed as Sclerophyllum coarctatum); and 'O. subulata' (to the genus Rhynchoryza).
$\textbf{c.} \ \ \text{For specimens of uncertain morphological identity, a determination of somatic chromosome numbers will be helpful in certain cases.}$
<b>d.</b> Annual and perennial growth habits are rather difficult to distinguish when the plants are grown under tropical conditions; the term "annual" refers to a primarily seed-propagated form while "perennial" refers to a taxon adapted to vegetative propagation by underground plant parts.

Appendix 2. A sample sheet from the IRRI varietal catalog (IRRI, 1970).

		7	/./	1/2	Z	Blode		/s/L	igule/	72		Culm		1.	Panie	le	G	ain	18/8/	¢//	Aw	1	77	1/2	1	17	Stem	boreri
/ 20 /	/ 0	/	1 4 /8	138	/ /	/0/.	1/	8//	13/	ø/	/	//	//	8	//	/4/	,	/ /	0/0/	\$ 18	///	\$ 18	10/3	12/	F /0	/	151	2/1
/ § /	1 3	1.	3/8/	0/	/	18/8	/ / 🗟	/_/	18/3	2/	/ /	/ / 🔅	1/2/2	*/	/ /8	13/	/	1	/8/	2/3/8	//	8/8/		2/0	18/	*/	\$/4	///
Name	/ 3 /	0 /.	9/1/1	/ 3	18	18/0/	8/8/	\$ /\$/	3/2/	\$/	/	2/8	8/8	15/	2/5/	1/ 1	/ 8	15	5/8	(3/§	3/8	18/5	/3/2	18	/\$/	§ / .	8/3	10/1
* /	19/	0/.	7/28/38	15	15	12/2/8	13/4	18/0	9/4/	5/6	?/8	18/1	E/E/	\$ /5	18/2	1 4	15	15/4	5/5/4	2/8/	198/	8/4/	5/4	12/	4 ×	/ 8	15/	4/4
1 2	3 4	5	6 7	8.	9	10 11 12	13 14	15 16	17 1	8 19	20	21 22	23 2	24 25	26 27	28	29	30 31	32 33	34 35	36 3	38 3	9 40	41 4:	2 43	44	45 4	647
7458 R1220110 x R. BERSANI	92- 5 4	8	2 30	51	1.4	2 3 3	1 16	0 1	0	67 15	5 1	6 4	1 1	6 3	3 ذ	10.7	3.8	3 0	1 0	0	0 0	2 1	1 3	43 2	118	5	3 (	6 4
6934 KL GOPHER	92- 7 9.	2	2 33	65	1.4	2 1 3	1 15	0 1	0 1	15 10	0 1	6 4	1 2	8 3	3 3	9.0	3.3	3 0	1 0	0	0 0	2	1 3	25 2	131	15	9	1 0
6955 RL GOPHER	92-7 9	2	2 27	65	1.9	2 1 3	1 19	0 1	0 10	07 15	5 1	6 4	1 2	5 3	3 3	8.3	3.9	3 0	1 0	0	0 0	2 :	1 2	30 2	1 23	16	11	4
4252 R-MOU-SHAN	21- 8 2	0	1 22	54	1.3	2 3 4	2 21	1 2	1 1.	20 20	2	5 4	1 2	6 1	3 2	8.3	2.0	3 0	1 2	0	2 0	2 1	1 3	20 1	411	4	4 (	6 7
4559 RU-DAU 3065	21- 8 2	0	1 23	43	1.2	2 3 3	1 11	0 1	0 1	(7 15	5 1	5 4	1 2	1 1	3 2	8.0	3.3	3 0	1 0	0	0 0	2 1	2	20 1	108		5 :	3
9247 KODJO LELE	44- 3 4	4	3 21	53	1.7	2 1 2	1 15	0 1	0 1	15 10	0 1	6 4	1 2	7 2	3 3	9.9	3.3	5 0	1 0	2 1	2 0	2 1	. 3	37 2	140	13	13	7
9909 RODJOLELE	44- 4	4	3 23	59	1.6	2 1 3	1 19	0 1	0 1	10 15	5 1	6 4			3 3	9.9		2 0	1 0	2 1	2 0	ě i	. 3	25 2	171			5
4439 RU-GG- SHLN	21-8 2		1 25	50	1.3	2 3 2	1 20			09 20					3 6	8.4		3 0			0 0		. 2	16 1	1 20	1	5 5	5 8
5037 KUGUE (8464799) 420	73- 8 9		1 20	48	1.4	1 3 3	1 10	0 1		06 10		é 4				10.1					0 0		2	27 2	117	6	34	7
379 KUJOFOISY	56- 1 5		1 25	49	.9	2 1 4	2 22	1 1		93 35				2 1		7.5			1 2		2 0			26 1	102	4	6	1
6173 MULLED LEAF I 1086	42-47 4		2 1 20	78	1.2	2 3 2	2 29	0 1		39 25		6 2		0 1							2 0			24 1	161		13	7
1710 ROLLER FORTUNA	92-5 9		1 20	48	1.9	2 3 2	1 18	0 1		67 25						10.2						2 1		28 2	112	12		
1711 ROLLER FORTUNA	92-5 9		1 20	53	1.7	2 3 2	1 16			67 20						10.4					2 0		. 3	31 1	112	20		5 7
7422 ROMANICO	92-5 7	5	2 25	34		2 3 3	1 12			70 6	вı	6 4	1 1	9 3	3 3	9.2	3.5	3 4			2 0		3		118			7 7
9280 HONCAROLO	41- 1		2 28	37	1.0	2 3 3	1 13	0 1		51 15	5 1	4 4	1 1	7 3	3 3	8.8	4.0	3 0	1 0		0 0		. 2	34 2	118			7 8
9301 KONCO	41- 1		2 22	39	.9	2 3 3	1 14	0 1		48 20		4 4		0 3	3 3	9.4	3.4	3 0			0 0		. 2	32 2	118	12		7 7
4440 ROD-HMA-BIR	21-8 2		2 21	49	1.1	2 3 2	1 16	0 1		85 20		4 4	1 2		3 3	8.0	3.6	2 0			0 0		. 3	22 2	125	5	3 7	
4441 KOU-HWA-BIR	21-8 2		2 22	43		2 3 2	1 12	0 1		20 10		6 4	1 2		3 3		3.3	3 0			0 0		. 2	24 2	1 20			8
4842 RUO-KIAN-HUAN	21-8 2		1 28		1.3	2 1 2				99 20				3 2		7.3			1 0		0 0			17 1	117	6	•	
7126 ROS SUCON 129	97-3 8		1 35	74	1.4	2 1 2	1 22	0 1		57 30	-		1 2						1 0		0 0		3	27 1	192	10	. 4	
3144 ROVER CELLA	92-5 4		2 20	50	1.2	2 3 3	1 14	0 1		71 15		5 4	1 2		3 3	8.3			1 0		0 0		3	28 2	110	5	2 1	
4436 RU- WUAN- SHUN	21-0 2		1 20	41		2 3 2	1 20	0 1		21 25		4 4	1 2		3 2		2.5	3 0			0 0		. 3	18 1	111	1		8
4839 RO- NUAN- SHUN 21-3	21-8 2		1 24	41	1.0	2 1 2	1 18	0 1		06 20		5 4	1 2		3 2	9.0	2.9				0 0			19 1	124	2	4	
313 RPA 3	73- 2 7		1 23	6.6		2 2 2 2 2 2 2 2 2	1 17		0 1					8 2		8.3			1 2		0 0				124		6	
9033 RP1 37-1	73- 6 7		1 22	56		2 3 2				4 20			1 2			8.6					0 0				146			. 3
314 RPI 51			1 20	10	1.3		2 13			93 20			1 2		3 2				1 0		0 0		3	20 1	161		3 1	٠.١
315 RPP 49	73-2 7		1 21	66	1.6		1 13			7 15				o i					1 2		0 0			23 2	112		6 1	
1647 H-PURPLE LEAF X GUIDRY SEL.	92- 5 9		1 22	4.5	1 4	1 3 3	1 12			0 15		6 4			3 2		2.6	3 0	1 2		0 0		3	21 2	112		5	6
1906 R PURPLE LEAF x 7/8 -3-10	73- 2 7		1 23	69	2.0	2 3 4	2 25	1 1	0 1			7 4			3 2	8.9	2.8	2 0			2 0		3	22 1	146	.,		5
310 RP 21 311 RP 33	73- 2 7		1 19	7 2			2 20	ii				6 4	1 3		3 2	8.9	3.0		1 0		2 0		3	20 1	146		7 1	4
311 KP 33 312 KP 41	73- 2 7		1 23	# 1			2 22		1 12					6 1	5 5		2.8				2 0	2 1	3	26 1	146	12	i	
8177 K15 4	21- 3 9		1 33	62		2 1 3									3 2	10.2					0 0	2 1		21 1	1 30	12	7 7	
8233 RTS 5	21- 3 9		1	61			1 22			31 30		6 3	1 2		3 2				2 0		0 0	2 1	3	16 1	1 38	7	2 6	
8234 RTS 12	21- 3 9		1 24	66		2 1 2	1 18	0 1		28 20		5 4	1 3		3 2				1 0		0 0	2 1	2	23 1	1 31	4	3 1	
8176 RTS 14	21- 3 9		1 29	73	1.3		1 19	0 1	0 13			6 4	1 3		3 2				1 0		0 0		3	16 1	1 48	4	, i	
8235 KIS 16	21- 3 9		1 24	60	1.3	2 1 2	1 21	o i		6 20		5 4	1 2		5 2				4 0		0 0		3	18 1	1 38	7	,	
8236 R15 23	21- 3 9		1 21		1.3	2 1 4	3 25	1 2		6 20		6 4	3 2						1 2		2 0		2		120		1	5
5414 RT 1006	80-1 8		1 22	53		2 3 3				5 20		0 4	1 2	9 1	3 2				1 0		0 0				125			- 1
3406 RUBIO	92- 5 7		1 18			2 3 2		0 1	0 11	1 10	1	5 4	1 2	0 1	3 2										110	13	4 3	.
				_				Testes			1	T T	TT-															_
1 2	3 1	4 5	6 7			10 11 12	13  14	13 16	[17]	10   19	20	21 22	123 2	4 25	26 27	28	29	30 31	32 33	34 35	36 37	138 3	7140	<b>41  42</b>	43	1 44	45 4	647

Appendix 3. Form for recording plant characteristics of O. sativa cultivars in the field.

# THE INTERNATIONAL RICE RESEARCH INSTITUTE

1.	NAME	CROP
2.	PLOT	NO ACC. NO
3.	SEEDL	ING HEIGHT (18 days after flushing) (cm
4.	,	BLADE (1st leaf below flag leaf on main culm)  Pubescence: glabrous, pubescent  Length:
_		Flag leaf (at full blooming): erect, intermediate, horizontal, descending
5.		SHEATH COLOR (base of plant) Surface: green, purple trace, purple, others
6.		E (1st leaf below flag leaf on main culm) Presence: present, absent Length: short, medium, long Color: colorless to green, purple, others
7.	COLLA	AR Color: colorless (white), green, purple, absent
8.	AURIC	CLES Color: colorless to green, purple, absent
9.		(on main culm at first flowering) Diameter: small, medium, large (mm Number: Angle: erect, intermediate, spreading Strength: brittle, lodged, weak, sturdy Color: (surfaces): green, gold, purple, others
		Length: (panicle base): (cm
10.	-	RESCENCE Stigma color: colorless (white and yellow), purple trace, purple, others
11.	. STER	ILE LEMMAS (tip grains near maturing) Color: colorless or white, straw, gold, red, purple, others
12.	. LEMM	1A AND PALEA (tip grains start maturing) Color: straw, gold (to brown), brown spots, brown furrows, red, purple black
13.	. AWN	(tip grains start maturing) Presence: absent, partly present, fully awned Apiculus: colorless (white) to straw, red, purple Color: straw, gold, red, purple
14	. SPIKE	ELET FERTILITY: highly sterile (under 50%). partly sterile (51 to 75% Fertilizers. (76 to 100%)
15	. PANIC	CLE
		Type: open, intermediate, compact, clustering Threshability: shattering, intermediate, tight Exsertion: enclosed, partly exserted, exserted
16	. LEAF	SENESCENCE AT MATURITY: quick, slow

17. MATURITY (days): 100, 100-115, 116-130, 131-145, 146-160, 161-175, 176-190,

191-205.

# Appendix 4

	ce Evaluation Experiment Record General Information	
	Experiment No	
Station		
Site (field no.)	Plot Nos	
Experimental design		
No. of replications		
No. of accessions	No. of control varieties	
Treatments:		
No. of levels/treatment		
Plot size:	Length of row	(m
	(cm) No. of seedlings per hill	
		_
	Transplanting date	
N	kg/ha as	
	kg/ha as	

Appendix 4 continued

kg/ha askg/ha as	
	lea /h
kg/ha at	kg/n
Texture	
Color	
Date or growth stage	
Date or growth stage	
Date or growth stage	
sures	
sures	
	kg/ha at

# B. Field Book (an example of screening for susceptibility to zinc deficiency)

Year		_ Crop			_					Experi	ment no.		_ Loca	tion	
Plot	Accession	Acc.	Seed			Tiller	Pt	Days t	o Leaf	discoloration	Lodging			Yield	
no.	name	no.	source	Tr.ª	Rep.	no.	ht. 90%	hd.	Matur.Score <sup>b</sup>	Stage <sup>b</sup>	%	Harvest	area	Actual	kg/ha
01	H-4 (check)			1	1										
02	CO 25			1	1										
03	Mahsuri			1	1										
04	Peta			1	1										
05	HR 35			1	1										
06	MI-48			1	1										
07	KU-10			1	1										
80	IR20			1	1										
09	C12			1	1										
10	E425			1	1										
11	C 12			2	1										
12	CO 25			2	1										
13	KU-10			2	1										
14	H-4 (check)			2	1										
	E425			2	1										
16	Mahsuri			2	1										
17	IR20			2	1										
18	HR 35			2	1										
19	Peta			2	1										
	MI-48			2	1										

<sup>&</sup>lt;sup>a</sup> Treatments refer to the application or no application of zinc. <sup>b</sup> In codes.

#### GLOSSARY

- **accession.** A variety or a strain or a bulk population registered at the national center and worth conservation. Two or more morphologically or ecologically different accessions may have the same name.
- **adaptability.** The ability to adapt to different environments by modifications in physiological responses.
- **adaptation.** The process of becoming suited to new or different environmental conditions or for particular functions.
- **adaptiveness.** Degree of being adapted to a certain environment or environments.
- **alleles.** One of two (or more) alternate forms of a gene located at a certain position (locus) on a particular chromosome or linkage group. When the number of alternate members exceeds two, the alleles form a multiple allelic series.
- **allelism.** The relationship between alleles in different parents. When the alleles of two parents belong to the same gene (locus), they are allelic; otherwise, non-allelic.
- **anisomeric genes.** Several genes that have one-directional and unequal effects. Their expressivity and heritability are intermediate between those of major genes and polygenes.
- biotic environment. The insect pests, plant pathogens, weeds, other crop plants, and small animals in the habitat comprise the biotic phase of environment.
- **bulk.** The growing and maintenance of genetically different plants in a population without separating them into pure lines or applying selection efforts.
- collection. A collected sample.
- **conserved stock.** An accession or its subdivisions (strains or pure lines) chosen for maintenance at a national or international center.
- cultivar. A cultivated variety; the international term for variety.
- **duplicate samples.** Collected samples from different sources which belong to the same variety as indicated by name, site of collection or origin, and morpho-agronomic characters.
- eco-strains. Strains within a variety that have developed physiological differences in response to repeated planting and selection in distinct environments.
- **edaphic factors.** Soil conditions such as alkalinity, extreme acidity, iron toxicity, salinity, and zinc deficiency, which adversely affect plant growth.
- **elite germ plasm.** Breeding stocks including cultivars of hybrid origin, breeding lines, hybrids of single or multiple crosses, bulk populations, and composite populations.
- **expressivity.** The degree of expression of a given gene. Genes that always produce the same phenotype have 100 percent expressivity.
- **gene.** The genetic unit controlling the inheritance of a character (trait). A character may be governed by one or several genes
- $\begin{tabular}{ll} \textbf{gene-pools.} \end{tabular} Desired genes or gene-complexes in genetically diverse populations. \\ \end{tabular}$
- **genetic base.** The amount of genetic variousness available to the breeder among widely grown cultivars in relation to the total variousness in a crop species. Cultivars with a broad genetic base can adapt more readily to changing

environments or selection efforts than can a narrowly based one.

**genetic composition.** The constitution and proportion of genetically different individuals in a population.

**genetic conservation.** Collection, maintenance, and preservation of all segments of germ plasm in a crop species and its wild relatives.

genetic diversity. Existing condition of being genetically different.

**genetic drift.** Changes in the gene frequencies of a population when the size of sample chosen for rejuvenation is small. Genetic drift leads to a loss of certain genotypes in the population.

**genetic potential.** The likelihood that the genetic material (improved or primitive) is capable of producing many new genotypic combinations from stored variousness by hybridization and subsequent segregation, recombination, and selection under varying pressures.

**genetic resources.** Germ plasm that includes the entire array of cultivars in the crop species, related wild species in the genus, and hybrids between the wild and cultivated species.

**genetic resources center.** An institution concerned with the overall conservation of a crop or crops.

**genetic stock.** A variety or strain known to carry one or more specific traits, though the genetic mechanism of such characteristics may not be so clearly understood as that of a genetic tester.

**genetic tester.** A pure line or hybrid known to carry one or more genes that distinctly express themselves in morphological or physiological features. The mode of inheritance of such traits is known by prior genetic experiments.

**genetic variability.** The ability to vary genetically.

genetic variation. The genetic process of varying.

**genetic variousness.** Diversity among individuals within a species or a smaller subdivision of the species due to differences in genetic constitution (genotype).

**genotype.** The genetic constitution or make-up of an individual organism; a group of individuals having a common or specified genetic make-up.

germ plasm. The sum total of genetic material in a species.

**heritability.** Proportion of the total phenotypic variance that is genetic for a given trait.

**heterogeneous.** Having different genotypes in a population. A heterogeneous population can have both homozygous and heterozygous individuals.

heterozygous. Hybrid for the different alleles of a gene or genes.

**homogeneous.** Uniform in appearance and similar in genetic composition due to descent from a common ancestor.

**homozygous.** Having one type of allele in both chromosomes for a given gene or genes.

**line.** A strain; a family derived from one plant.

off-types. Plants that differ in morpho-agronomic characters from the majority or representative plants of a variety; admixtures in a field; obvious contaminants such as tall plants in a semidwarf cultivar or vice

**oligogenic gene.** A major gene of good expressivity and high heritability. Mendelian analysis of discontinuous traits generally involves major genes.

- **phenotype.** The appearance of an individual produced by the genotype in interaction with a given environment.
- **physical environment.** The climatic factors, the soil, and free water surrounding the rice plant comprise the physical environment.
- **polygenes.** Numerous genes that contribute to the continuous variation in a quantitative trait, each having small and equal effect.
- **polymorphism.** Simultaneous and regular occurrence in an essentially homogeneous population of two or more types of variants that are genetically related.
- **population.** A group of individuals (plants) within a species or a variety that are found at one site or field. Plants in the population may or may not be genetically alike.
- **primitive forms.** Plants having primitive features such as pigmented plant parts, long awns, lax panicles, extreme shattering, and perennial growth habit.
- **pure line.** A line that has been made almost completely homozygous by repeated self-pollination and selection of a specific type (or by the removal of off-types).
- **regional conservation center.** A national or international center located in a broad geographic area that assumes the responsibility for conserving germ plasm in that area through collection, rejuvenation, and storage. The regional center also handles the distribution of conserved stocks. It cooperates with the genetic resources center in overall preservation of a crop or crops.
- **strain.** A group of plants within a variety that share common, recognizable features in morpho-agronomic characters, physiological differences, or reactions to specific disease pathogens or insect pests. A strain may include several lines with similar features.
- sample. Individuals taken from a population to represent it.
- variants. Related members of a population showing small but distinct genetic differences in one or two features.
- varietal diversity. The extent of dissimilar varieties growing in an area.
- variety. A group of cultivated plants within a species which is distinguished from another variety (group) by any characters (morphologic, physiological, biochemical, or other) of significance to agriculture and which, when reproduced, retains its distinguishing characteristics. A variety may be derived from several pure lines which have many common features and are reasonably uniform in appearance (but not necessarily genetically pure).
- working collection. A sizeable collection of evaluated accessions that are adequately stored, documented, and available for immediate use.
- world collection. A comprehensive collection of samples from different geographic areas of the world held in storage for preservation. Only segments of the world collection are of immediate practical value and thus find their way into the working collection.

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